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SKELETAL MUSCLE FATTY ACID COMPOSITION OF WILD
UNGULATES AND FORENSIC APPLICATIONS

by



ROBERT ALLAN McCLYMONT

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH
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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research, for acceptance, a thesis entitled "Skeletal Muscle Fatty Acid Composition of Wild Ungulates and Forensic Applications", submitted by Robert Allan McClymont in partial fulfillment of the requirements for the degree of Master of Science.
in Animal Biochemistry.

ABSTRACT

Stepwise discriminant function analysis was used to compare longissimus dorsi muscle samples from elk, moose, white-tailed deer, mule deer, antelope and bighorn sheep using triacylglycerol(TG) and phospholipid(PL) fatty acid values. Each species was significantly different ($P < .01$) from each of the others except for white-tailed deer and mule deer triacylglycerols which were significantly different at $P < .05$. Elk in particular had a distinctive fatty acid pattern. Diet and time of year appeared to have little influence on the fatty acid patterns.

Longissimus dorsi samples for each of elk, moose and white-tailed deer were compared by age and by sex. Changes in fatty acid values with age were not consistent among the three species but some differences apparently due to age were noted. For each species some fatty acids tended to be different between males and females but in all cases the two groups had overlapping ranges.

The effect of anatomical location on the values for the major individual TG and PL fatty acids was examined in a number of individuals. PL fatty acid values appeared to be little affected but there was a tendency for TG fatty acids to be less saturated in muscle samples from more peripheral muscles.

Fatty acid values of samples from a variety of muscles were used to derive classification functions for the classification of samples to species and for elk, moose and white-tailed deer samples to age and to sex as well. The overall success rates for the various classifications varied from 77% to 100%. After the initial classification of samples to species each species was then compared with only those

species that had samples misclassified into that species. Through this method it was possible in some cases to reduce or eliminate misclassifications. Trends observed for values of some fatty acids led to a comparison of species using the ratio (oleic/myristoleic + palmitoleic + oleic) 100%. Elk samples were found to have values between 50 and 80% except for two calves which had values greater than 80%. All samples from the other species except for two moose samples had values greater than 80%.

Cooking of meat samples to internal temperatures of 61 C or 77 C did not appear to affect fatty acid composition.

It appears that while fatty acid analysis by itself may not always provide a clear-cut classification of a meat sample as to species, age or sex it is at least a useful adjunct to other classification techniques.

Various analytical techniques applicable to the identification of wildlife tissues are reviewed in this thesis.

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REVIEW OF FORENSIC TECHNIQUES

Introduction

In Canada, Provincial Fish and Wildlife Branches and the Canadian Wildlife Service are charged with the responsibility of conservation and management of fish and wildlife resources. Their efforts are divided among three basic activities: research, management and enforcement. Research is aimed primarily at keeping management techniques up to date while little research emphasis is placed on enforcement problems. This is rather unfortunate because "Without adequate law enforcement, the finest research and management programs would have little meaning." (Morse, 1971). In the future, as public demands on wildlife resources increase, it will become increasingly more important to have efficient control over the use of these resources. Among other things, this will depend upon the immediate development of forensic science through research using new as well as existing analytical techniques.

One of the biggest hurdles to establishing a good game management program is the prevention of poaching. The seriousness of poaching to game management can be illustrated by a couple of examples. Beattie et al. (1977) reported that a simulation study by Vilkitis estimated the illegal kill of big game animals during the 1967 closed season in Idaho to be about 2400 animals. They also report that Vilkitis repeated his simulation of big game poaching for the 1970-71 Maine closed season and from this he estimated that during the five month period some 18,000 deer were taken. In general, it is usually impossible to convict violators unless they are caught in the act or with obvious evidence of their guilt. However, because of recent advances in various fields of

science this situation may soon be remedied and it will be possible to detect breaches of the law even when the violator has attempted to conceal his guilt. Modern forensic techniques have mostly been applied to investigations of criminal code violations but some may be equally applied to wildlife cases (Oates et al., 1974; Flynn and Franzmann, 1977; Hoekstra and Carr, 1977). Since the prime concern of wildlife agencies is the conservation of wildlife, it is hoped that public awareness of the modern forensic techniques at the disposal of these agencies will serve as a strong deterrent to potential violators rather than merely as a tool to convict more violators.

During investigations of game act violations, some of the important factors which must often be determined are species, sex, age, time of death and geographic origin of an animal. Techniques which have or could be used to answer such questions involve the physical examination of bones and hair and the chemical characterization of hair, meat, blood and other tissues.

An ideal technique is one that is simple, fast, repeatable, based on thoroughly established chemical and/or physical principles, and provides a maximum of information about a sample. One technique that perhaps most closely approaches such requirements is electrophoresis of tissue proteins. However, it would be useful in certain situations to verify electrophoretic results using additional techniques, and at other times alternate techniques might have to be used if the proteins have been severely damaged as in cooking.

An investigation of tissue fatty acid composition was undertaken to determine its' usefulness as an alternate or supportive technique to electrophoresis. The results and possible forensic applications of this study will be the subject of this thesis. First, however, the

author would like to review forensic and other scientific techniques which have been or could possibly be applied to wildlife forensic investigations. The presentation of these techniques will be under the headings of the specific problems to which they may be applied.

Species Determination

The identification of species on the basis of the examination of only a portion of a carcass is a major concern of the wildlife forensic scientist. At times it is possible to identify the species of origin of viscera from the physical characteristics of various parts; for example the nature of the surface of the kidney, the presence or absence of a gall bladder, the structure of the larynx, or the absolute and relative sizes of various structures (Sisson and Grossman, 1953). An examination of the parasite species present in the viscera may furnish a key to its species of origin on the basis of the host specificity of the parasites recovered.

The physical characteristics of fecal material can be used in some cases to identify the species of origin. The measurement of fecal pH has also been suggested as a technique for the identification of species origin (Howard, 1967; Nagy and Gilbert, 1968; Krausman et al., 1974). There are a number of factors, however, that could cause intraspecific variations in fecal pH so the usefulness of this technique would need to be evaluated on a regional basis. Determination of parasite eggs and larvae in feces can also furnish a key as to species of origin.

A more commonly encountered situation is the need to identify a dressed carcass which may, in many cases, have already been packaged.

Archeologists identify species of skeletal remains on the basis of size and shape of various bones (Gilbert, 1973; Hildebrand, 1955; Schmid, 1972). However, bones are frequently unavailable or may be of limited taxonomic value because of the manner in which the carcass has been processed. To accurately identify bones it is essential that the investigator have an adequate reference skeletal collection of different age and sex classes for each species to be considered.

In many instances hair has proven to be useful for species determination. It is usually possible to recover samples of hair from instruments used in the processing and handling of animals or from the meat itself. Physical differences in hair such as size, color pattern and cuticular scale pattern have been used as a key to species identification (Moore et al., 1974). Because of possible variations in the cuticular pattern along the length of an individual hair, on hair from different parts of the body and on hair from different individuals of the same species, it may be unrealistic to expect to differentiate closely related species using this technique.

Chromosome number and structure can be used to distinguish between some species (Gustavsson and Sundt, 1968; Hsu and Benirschke, 1969). Care must be taken however, as different species may have the same basic chromosome number and there can be some intraspecific variation in number. A further consideration in chromosome analysis is that the material to be examined must be fresh and contain a sufficient number of dividing nuclei. Culturing techniques may be required to obtain sufficient numbers of suitable nuclei. Culturing cells is a difficult technique and obtaining viable cultures from post-mortem tissues depends on the type of tissue, length of time post mortem and

environmental conditions during the post-mortem period (Ludwig and Titus, 1972). Tissues would probably have to be obtained within 12 to 18 hours post-mortem.

Although it has not been used for species identification in a forensic context, interspecific differences in element composition have been demonstrated in comparisons of waterfowl feathers (Kelsall et al., 1975) and in fish tissues (Calaprice, 1970). Because of modifying factors such as diet, health, and geographic location this technique will probably be of limited value for species determination of game animals.

The most commonly reported techniques used for species identification involve the determination of the immunological reactivity of proteins from tissues and body fluids. In the past the precipitin test (Keiss and Morrison, 1956) was widely used. Some workers felt that this technique was not specific or sensitive enough and other tests have been applied such as passive hemagglutination inhibition (Tempelis and Rodrick, 1973), immunodiffusion (Fugate and Penn, 1971), immunoelectrophoresis (Oates et al., 1974; Oates and Weigel, 1976) and radio-immunoassay (Croonquist, 1977). The determination of species by immunologically testing blood and meat has been accepted as evidence in court. A number of references on this subject are discussed by Oates et al. (1974). In these cases the antibodies used have been for native antigens of various animal species. Analyzing the tissue in question for antibodies against certain parasites and diseases, may also furnish some evidence as to species (Werrett et al., 1976). The success of this latter technique would depend on preservation of the activity of antibodies in the tissue. The fact that antibody

techniques are not used more widely is likely due in large part to the difficulties associated with the production of discriminating antibodies and the variability in the specificity of antibodies produced from one time to the next.

Seed lectins from certain plant species have been shown to have differing specificities for agglutination of blood which make it possible to differentiate certain animal species (Bhatia, 1974; Bird, 1954) but to the author's knowledge this has not been applied to the forensic determination of species. Other blood typing systems might also furnish some clues for species determination.

Chromatography and electrophoresis of tissue proteins have been widely used for species identification. Staining for total proteins or for specific proteins from fresh, frozen and cooked muscle (Mackie, 1969; Dilworth and McKenzie, 1970) has been used. Hair and feather proteins (Brush, 1976; Darskus and Gillespie, 1971) and blood proteins (Bunch et al., 1976) have also been examined electrophoretically. The criteria used for identification are the number of protein bands that are formed and the migration distance and staining intensity of these bands. These band characteristics for muscle and blood samples can be influenced by a number of variables such as the specific tissue used, individual variation, physiological condition of the animal at the time of death and postmortem conditions (Markert and Møller, 1959; Aberle and Merkel, 1966; Hay et al., 1973; Miller et al., 1965; Grunbaum, 1977). The investigator must be very conscious of these factors when interpreting the results of such analyses.

In addition to blood protein differences observed using electrophoretic techniques other interspecific differences in blood have been observed. A comparison of pronghorn antelope with four deer species

showed differences in the concentrations of some serum proteins between antelope and the cervids (Dhindsa et al., 1975). Concentrations of hemoglobin (Herin, 1968) and carotenoids (Knight, 1969) have also been shown to differ between some species. The usefulness of concentrations of various materials in blood is limited for distinguishing species because of the tremendous animal-to-animal variation that may occur.

Reichert and Brown (1909) demonstrated that hemoglobin crystal patterns differ between species. Use of this technique for species determination has been reported by several authors. Among these is a report by Winter and Honess (1952) wherein hemoglobin crystal structure was used as evidence in a wildlife court case. Washino and Else (1972) used hemoglobin crystal patterns to determine on which species hematophagous arthropods had been feeding. The presence or absence of sickled erythrocytes may also furnish a clue as to the species of origin. In a survey of the Artiodactyla, Butcher and Hawkey (1977) observed sickling in 9 out of 13 species of deer and in 2 out of 12 bovid species examined.

Sex Determination

The removal of obvious evidence of sex is perhaps one of the more common violations of the game act. This is often done out of ignorance of the law, but during seasons when only animals of a specific sex may be taken it may be done intentionally by the hunter. One method of determining sex in such cases is to examine the structure of the pelvic bones. From an examination of the pelvic bones from black-tailed and white-tailed deer Taber (1956) concluded that there were three characteristics which indicate sex:

1. outline of the pubic symphysis
 male - thick and blunt
 female - more slender and pointed
2. suspensory tuberosity on the inter-ischial bone
 male - present
 female - absent
3. relative length of the pubis (pubis length/pubis symphysis depth) greater in females than males.

Evidence of the sex of a carcass may also be apparent by the absence or presence of the pizzelle eye (part of the crural attachment of the penis), cod fat (fat from scrotal region of males), and udder fat, and in a split carcass by the depth and length of the exposed surface of the gracilis muscle adjacent to the aitch-bone (Denney, 1965).

Microscopic examination of tissues can also be used to determine sex. Dividing cells from cell cultures can often be examined for sex chromosomes. However, viable cells are required for this technique. Also, blood polymorphonuclear leukocytes display sexual dimorphism in their structure (Davidson, 1966). It may also be possible to observe this dimorphism in cells from the red marrow of ribs and the femur which are sites of blood cell formation. A comparable structure to the drumstick-like nuclear appendage of leukocytes seen in blood from females occurs in tissue cells. These structures are called Barr bodies or X-chromatin and are seen as dark staining bodies typically lying at the inner edge of the nuclear membrane. Moore (1966) cites the results of investigations on X-chromatin in tissues from members of the artiodactyls. On the basis of the limited amount of work done on artiodactyla it appears that in cells of most non-neural tissue from these animals sexual dimorphism is obscured by excessive amounts of

coarse autosomal chromatin, but in suitable tissues the X-chromatin can be observed for some time after death of the animal. The percentage of neural cells with visible X-chromatin has been used in wildlife law enforcement cases for determining sex (Hoekstra and Carr, 1977). The presence of these structures ('drumsticks' and 'Barr bodies') only indicates femaleness and can give no indication as to the maleness of a tissue. A histological technique in which the Y-chromatin is stained with a fluorescent dye has recently been developed for identifying maleness of cells. The technique has been applied in human forensic medicine (Vakil et al., 1973) and is now being developed for wildlife identification (Hoekstra and Carr, 1977). Hoekstra and Carr (1977) mention that testing for the H-Y antigen may be useful for sex determination of tissues when post-mortem decay has made it impractical to examine for X- and Y-chromatin.

Sex related differences in the mobility and density of some bands seen in electrophoresis of deer and avian blood proteins have been reported (van Tets and Cowan, 1966; Moore, 1945) but have not been applied to forensic science.

Sweet and Elvins (1976) using crossed electroimmunodiffusion reported finding significant differences between human females and males for quantities of various antigens in blood stains. This type of study has not been extended to wildlife species.

Geographic Origin

Because wildlife is managed on a regional basis the question of where an animal was killed is sometimes encountered. There are several reports in the literature of intraspecific regional or population

differences in electrophoretic properties of blood proteins (Cowan and Johnston, 1962; Cummings, 1970; Harris et al., 1973). Such regional differences have also been reported in the mineral composition for hair, feathers and fish tissue (Franzmann et al., 1977; Kelsall and Burton, 1977; Calaprice, 1971). Regional differences in element composition are likely the result of differences in dietary uptake although factors such as physiological conditions, health and environmental factors will have some effect. Differences in elemental composition may prove to be of forensic value when applied to animals known to occupy distinct areas but should be used with caution when dealing with species such as deer, as individuals may move long distances.

Time of Death

The question 'When did death occur?' is one that needs to be answered in cases involving suspected night hunting and hunting out of season. One method which has been suggested for estimating time of death in humans is measurement of potassium levels in the vitreous humour (Adjutantis and Coutselinis, 1972). To reduce the effect of internal variations these authors measured the rate of change of potassium levels and extrapolated this to determine zero time of change. Ambient temperature has been found to have no significant effect on the increase in potassium in the vitreous humour of human and cow eyes after death (Sturner and Gantner, 1964). Exposure of eyes to temperature extremes such as occurs with freezing or burning might result in unusual changes in potassium levels but was not tested for by these researchers. Measurement of potassium levels is one method currently being actively researched for estimating time of death in

humans but to the author's knowledge has not been applied to wildlife forensic work. Gill and O'Meara (1965) tested the reliability of a number of procedures for estimating time of death in white-tailed deer. Eye appearance, pupil diameter, carcass temperature and rigor-mortis were found to be the most useful characters. It is necessary for the investigator to know or make knowledgeable assumptions of post-mortem conditions in order to evaluate the reliability of these characters (Gill and O'Meara, 1965). In the above study the criteria used were found to be most reliable for deer dead less than 24 hours. Fish and Game officers in Maine and Virginia are being trained to use these criteria for estimating time of death and it has been used successfully in court. Brunetti et al. (1977) have suggested that the presence of whole red blood cells and evaluation of bacteria levels may offer some clues as to how long a piece of meat has been aging. There is some degree of variability in results obtained by the above techniques but often investigations require only a rough estimate of the time of death.

Techniques suitable for longer time periods are also necessary. Two independent studies on electrophoresis of deer blood proteins reported that albumin showed significant quantitative differences related to time of year (Cummings, 1970; van Tets and Cowan, 1966). Sibley and Johnsgard (1959) reported quantitative differences related to season for avian serum proteins. Seasonal variations have also been observed in the elemental composition of hair from cattle and moose (Hidioglou and Spurr, 1975; Franzmann et al., 1975). Tests such as these which rely on quantitative differences are subject to severe criticism on the basis of premortem individual variations that can exist and also because of the influence of differences in postmortem environmental factors. The establishment of 'normal' ranges for values and

elucidation of the causes of variation would add greatly to the usefulness of these quantitative techniques.

Information on what time of year an animal died may be provided by examination of the color pattern of hair or feathers (Brunetti et al., 1977) and by the degree of wear. If teeth are available, a histological examination of a section may provide an indication of what time of year the animal died (Bourque et al., 1978).

Some work has also been done on estimating bloodstain age. Kind and Watson (1973) compared the visible absorption spectra of human bloodstains up to 15 years old and found that the spectra varied in a regular manner with age. Cummings (1970), using starch gel electrophoresis, correlated an observed loss of pre- and post-albumin protein fractions from samples of deer bloodstains with age of the stains. This correlation was limited to stains less than two months old.

The length of time that meat has been in storage is another question which may need to be answered in some court cases. Studies on the effect of cold storage on fish muscle have indicated that a decrease in the solubility of myosin (Connell, 1962) and actomyosin (Love, 1962) is a measure of length of cold storage. Awad et al. (1968) reported a steady decrease in the solubility of both myofibrillar and sarcoplasmic proteins of bovine muscle during storage for 8 weeks at -4 C. Love and MacKay (1962) felt that determination of muscle cell fragility had several advantages compared with soluble protein determinations. Degree of freezer burn, depth of freeze-drying, color, etc. are used by the State Crime Lab in Madison, Wisconsin for determining length of storage of meat (Wagner, 1977). A great deal of caution must be exercised in interpreting these latter criteria as they may be greatly influenced by variations in storage conditions.

It may on occasion be necessary to determine if a particular meat sample is fresh or has been brought out of cold storage. Buchanan (1971) has described a simple technique based on the loss of the water holding capacity of meat after freezing, however this quality is greatly affected by the pH of the muscle and other variables. The presence of whole red blood cells is a good sign of freshness as freezing will cause hemolysis (Brunette et al., 1977). Electrophoresis may also be useful in determining if meat has been previously frozen. Escoubas et al. (1975) found that the number of lactate dehydrogenase bands in bovine muscle increased after the muscle had been frozen. Similar results have been reported for malate dehydrogenase by Blonde et al. (1967).

Tissue Matching

Another problem that arises for the wildlife forensic scientist is that of determining whether or not various tissues came from the same or different animals. Physical matching of portions of animals has been successfully employed in solving some cases (Wishart, 1977). Identification of muscles and bones present may provide information on how many animals are involved (Brunetti et al., 1977). The size and degree of ossification of bones may also serve as useful clues (Knight, 1966). Jones (1975) and Burnham et al. (1976) suggest that it may be possible to distinguish hair and bones from different individuals using photoluminescence. Burnham et al. (1976) state that bones from different individuals can usually be distinguished quite successfully by saturating the bones with a fluorescent dye solution, drying them, and examining the color pattern produced with an ultra-violet light source. The bones from different individuals will

apparently have characteristic color patterns which are dependent on the structure of the Haversian canal system. A court case is pending in Montana in which fluoride levels have been used to match vertebrae from the front and hind halves of an illegally killed bighorn sheep (Palmisciano, pers. comm.). Jones (1975) found that when hair proteins were made to phosphoresce by excitation with ultraviolet light, there were differences in the spectra and decay times for hair from different individuals. The number of donors was limited however, and he felt that for a large population this technique alone would be insufficient to individualize a hair sample. Perkons and Jervis (1962) had previously suggested that it might be possible to distinguish hair from different persons using neutron activation to make qualitative and quantitative determinations of elements present in hair. Flynn and Franzmann (1977) have examined the elemental composition of moose hair to determine if separate samples came from the same animal. Such analysis has resulted in charges being dropped in some cases and in suspects admitting their guilt in other cases, but as far as this author knows, Flynn and Franzmann's results have not yet been accepted as evidence by the court. Elemental analysis of human hair has been accepted as evidence in felony cases in Canadian courts. Recent research on electrophoresis of hair proteins has shown that it may be possible to use this technique as a means of identification of individual humans (Lee et al., 1978; Twibell and Whitehead, 1978). In a study of human bloodstains Sweet and Elvins (1976) were able to individualize each of the ten samples they examined using crossed electroimmunodiffusion to determine the quantities of various proteins present. They felt that at present this technique is unusable for large populations because of the lack of knowledge as to the normal ranges of concentration of these

proteins.

Determination of blood groups and/or isozyme types could be used to determine that a sample did not come from a specific animal or to determine the degree of probability that separate samples did come from a specific animal. The latter circumstance would require a knowledge of the frequency of each protein type in the population under consideration. This type of information is also necessary with many of the other techniques that have been considered in this review. In general, the methods discussed above could be more readily used to determine that tissue samples came from different animals than to prove that they were derived from the same animal.

Conclusion

A variety of techniques have been presented which may be of use to wildlife enforcement agencies as forensic tools. Many of these techniques are still at a theoretical or at a research level of development while a few are already in use. This review is not comprehensive; some theoretically possible techniques and some techniques employed in human forensic work have not been presented. Also, there are some problems which may be encountered in wildlife enforcement that have not been dealt with. It is quite apparent that the field of wildlife forensic science is in its infancy and that a great deal of research and cooperative effort will be required for it to approach its potential.

The subject of this thesis is the determination of the fatty acid composition of tissues from a variety of wild ungulates native to Canada and the assessment of the usefulness of this data as a forensic tool.

FATTY ACID COMPOSITION OF MUSCLE

INTRODUCTION

It has been known for some time that there are variations in tissue fatty acid composition among species. For example, lipid from ruminants contains much smaller proportions of the polyunsaturated fatty acids than does lipid from monogastric animals. This is largely due to the ability of rumen microorganisms to readily hydrogenate dietary polyunsaturated fatty acids to monoenoic or saturated fatty acids. The microorganisms in the rumen are also responsible for the production of small amounts of odd chain and branched chain fatty acids which are therefore found in higher concentrations in tissue from ruminants than monogastric animals (Hubbard and Pocklington, 1968). Therefore, it is often possible to determine if a sample of meat originated from a ruminant or from a monogastric animal. The fatty acid composition of tissues from different species of ruminants, however, tend to be similar. The first chapter of this thesis will examine the differences in fatty acid pattern of l. dorsi muscle among elk, moose, white-tailed deer, mule deer, antelope and bighorn sheep.

In addition to interspecific differences in fatty acid composition some studies on domestic species have indicated that age and sex have some influence on fatty acid composition. The effects of these two variables on the fatty acid composition of longissimus dorsi (l. dorsi) muscle from elk, moose and white-tailed deer are examined in Chapter 2.

Fatty acid composition of muscle has been reported to vary with the anatomical location of the muscle. The effect of this variable was examined in a number of individuals and the results are presented in

Chapter 3.

The underlying motive for determining the fatty acid composition of muscle from these species of wild ruminants was to see if this data would be of some forensic value. Chapter 4 presents the results of the species classification of a variety of muscles from the six wildlife species mentioned above. Chapter 5 deals with the age and sex classification of muscle samples from elk, moose and white-tailed deer. The effect of cooking on fatty acid composition of muscle is reported in Chapter 6.

INTRODUCTION

A considerable number of authors have reported the fatty acid composition of various tissues from domestic animals used for meat production (eg. sheep and beef) but only a few have reported the fatty acid composition of tissues from wild ruminants native to North America (Garton and Duncan, 1971; Garton et al., 1971). In this chapter the fatty acid composition of the longissimus dorsi muscle (l. dorsi) from elk (Cervus elaphus), moose (Alces alces), white-tailed deer (Odocoileus virginianus), mule deer (Odocoileus hemionus), antelope (Antilocapra americana), and bighorn sheep (Ovis canadensis) are examined.

EXPERIMENTAL

The muscle samples were obtained from animals killed by hunters, from a population reduction program at Elk Island National Park and from road-killed animals. Appendix 1 lists details regarding data of collection, species, age, sex, anatomical site and fatty acid composition for each sample. Most samples were from the l. dorsi from the lumbar region. Several other muscles from a number of individuals were also analysed as outlined in Chapter 3.

After collection, the samples were stored at -16 C until they were prepared for analysis. To facilitate adequate subsampling and the extraction of lipids the muscle samples were freeze-dried and then ground in a homogenizer (Janke and Kunkel DG IKA WERK Stanfen:Breisgau Type A1051). When whole muscles were studied the larger ones were first ground in a meat grinder and subsampled prior to freeze drying.

All solvents used in sample preparation were purified by distillation

Lipids were extracted from the freeze-dried muscle by a modification of the method of Folch et al. (1957). A 2.0 g sample of muscle was extracted for 3 to 5 min with 20 ml chloroform-methanol (2:1) in a Virtis homogenizer (Virtis Company, Yonkers, N.Y., U.S.A.). The resultant slurry was filtered by vacuum through Whatman No. 2 filter paper and the residue extracted with a second 20 ml volume of chloroform-methanol (2:1). The slurry was filtered as before and the total filtrate evaporated under an atmosphere of nitrogen to a volume of about 5 ml. About 0.25 ml of this solution was applied as a straight thin line across half of a commercially prepared 0.25 mm thick silica gel G thin-layer chromatography plate (Macherey-Nagel and Co.; Fisher Scientific Co. Ltd., Edmonton, Alta., Canada) about 25 mm from the bottom and 12 mm from the edge. Two samples, approximately 25 mm apart, were run on each plate. The plate was placed in a developing tank containing enough solvent (diethyl ether:benzene:ethanol:acetic acid, 40:50:2:0.2) to cover about the first 6 mm of the plate and developed until the solvent had moved half-way up the plate. The plate was then removed, air-dried and placed in a second tank which contained diethyl ether:hexane:acetic acid(25:75:1) and left until the solvent reached the top of the plate. The plate was removed, air-dried, and sprayed (except for the area of sample application) with 0.1% 2,7-dichloro-fluorescein in 98% ethanol. The lipid classes were visualized by viewing the plate under short wave UV light (UVSL.25; Ultra-Violet Products Inc.; San Gabriel, Calif., U.S.A.). The triacylglycerol (TG) and phospholipid (PL) bands were scraped from the plate into methylation tubes (20 x 150 mm pyrex culture tubes). A 10 to 15 ml volume of boron trifluoride solution (10% BF_3 in methanol:pentane:methanol, 35:20:45)

was added to each tube and the tubes sealed with teflon-lined screw caps. The tubes were heated in a boiling water bath for 40 to 60 min with occasional shaking. After cooling a tube to room temperature, 5 ml distilled water was added followed by 10 ml pentane and the tube was gently shaken. Upon standing for a couple of minutes the solution became biphasic with the upper (pentane) layer containing the fatty acid methyl esters. To purify the methyl esters the pentane layer was removed with a Pasteur pipet, reduced to less than 1 ml under an atmosphere of nitrogen and applied to a thin-layer plate as described above with the exception that four samples were applied to each plate. The plate was placed in a tank containing benzene and developed until the solvent reached the top of the plate. The methyl esters were located by observing the plate under UV light, scraped into a funnel containing a plug of glass wool and extracted from the silica gel by passing about 15 ml chloroform-methanol (2:1) through the funnel. The chloroform-methanol solution was then evaporated to dryness under an atmosphere of nitrogen and the methyl ester residue dissolved in a small volume of pentane and stored in a refrigerator. Prior to analysis by gas chromatography the pentane was evaporated and replaced with about 0.1 ml of carbon disulfide. About 1.5 μ l of the carbon disulfide solution was injected into a Bendix 2500 gas chromatograph equipped with flame ionization detectors (Bendix Corporation, Process Instruments Division, Ronceverte, W. Va., U.S.A.). Separation of fatty acid methyl esters was done in a glass column 3.7 m long, 3 mm internal diameter, packed with 10% Silar 5-CP on acid washed 100/120 mesh Chromasorb W (Applied Science Laboratories Inc.; State College, Pa., U.S.A.). Oven temperature was maintained at 225 C. Peak integration values were obtained with an Autolab Minigrator (Spectra Physics, Technical Market-

ing Associates; Mississauga, Ont., Canada) and the peaks were reproduced on a Fisher Recordall Series 5000 (Fisher Scientific Co. Ltd., Edmonton, Alta., Canada). Tentative peak identifications were made by comparing retention times of unknown methyl esters to those obtained for mixtures of known fatty acid methyl esters (Supelco Inc.; Bellefont, Pa., U.S.A., and Applied Science Laboratories; State College, Pa., U.S.A.). For each injectate the relative amount of each fatty acid, measured as its methyl ester, was determined by summing the integration values for all peaks and expressing each as a percent of the total.

Throughout this thesis the fatty acids will be indicated by the general shorthand form of XX:X, where XX indicates the number of carbon atoms in the molecule and X after the colon indicates the number of double bonds in the molecule. The animal species will frequently be represented by the following abbreviations: WTD - white-tailed deer, MD - mule deer, Ant - pronghorn antelope and BHS - bighorn sheep.

The fatty acid patterns of l. dorsi samples were compared by species using a SPSS discriminant function program as outlined by Klecka (1975). Stepwise discriminant analysis is designed to select and weight a set of variables such that differences between the groups are maximized. In the present case individual fatty acids were the variables used. At each step the variable which would increase Rao's V (a generalized measure of group separation) the most was added to the set of discriminating variables. The selection of variables was continued until the addition of the next variable would result in an F ratio (a measure of the additional power of discrimination introduced by the variable) of less than 1.0. The selected variables were then weighted and combined to form canonical variates such that the differences between mean scores

for the groups were maximized. The first variate is the single combination of weighted variables that gives the maximum separation of the groups relative to variability within the groups. The second variate is a combination of weighted variables uncorrelated with the first that gives maximum separation of the groups. The maximum number of variates that can be derived is equal to the smaller of either the number of variables or one less than the number of groups being discriminated. The F values in the tables of pair-wise comparisons of species represent the significance of the separation of the group centroids. The discriminating power of the variate(s) is indicated by Wilks' lambda - the smaller the value, the greater the discriminating power. Canonical correlation values represent the relative ability of a variate to separate the groups. The absolute values of the standardized canonical variate coefficients represent the relative contribution of the respective variable to the variate. Each sample is plotted on a 'map' with the scores for the first two variates as the axes. This display makes it easy to see the distribution of the samples and the degree of overlap and association of the species.

RESULTS AND DISCUSSION

The average concentration of each of the major fatty acids for the l. dorsi samples is shown in Table 1 for TG and Table 2 for PL. Values for individual muscles are tabulated in Appendix 1. There was little apparent variation between species in muscle PL fatty acid patterns. There is only a single report in the literature, providing values for PL fatty acids of muscle for wildlife species examined in the present study. That report is restricted to moose (Tanhuanpää and Pulliainen,

Table 1. Relative amounts of the major triacylglycerol fatty acids of longissimus dorsi muscle from some Alberta wild ruminants.

	Elk (18) ¹	Moose (23)	White-tailed deer (20)
Fatty Acid			
14:0	6.5 ± 2.1 (2.8-11.6) ²	1.2 ± 0.6 (0.5- 2.6)	1.3 ± 0.7 (0.3- 2.7)
14:1	2.7 ± 1.2 (0.3- 5.0)	tr	tr
16:0	35.6 ± 5.0 (25.9-43.7)	19.4 ± 2.5 (15.5-23.5)	22.1 ± 3.5 (17.6-30.6)
16:1	14.3 ± 5.0 (2.7-20.6)	2.5 ± 0.9 (1.5- 5.0)	2.3 ± 0.8 (1.4- 4.1)
18:0	10.2 ± 6.1 (5.4-27.6)	24.3 ± 4.5 (11.7-33.6)	28.3 ± 4.8 (19.8-40.8)
18:1	23.7 ± 3.2 (17.6-29.0)	42.8 ± 5.7 (31.0-56.2)	37.3 ± 6.2 (24.4-48.9)
18:2	2.9 ± 1.5 (1.2- 5.9)	3.6 ± 0.9 (2.0- 5.2)	3.3 ± 1.6 (1.9- 9.4)
18:3	tr ³ (0.1- 1.2)	1.0 ± 0.4 (0.3- 1.7)	1.0 ± 0.4 (0.2- 1.8)
20:3	tr (0 - 0.1)	tr (0 - 0.1)	tr (0 - 0.1)
20:4	tr (0 - 1.1)	tr (0 - 0.3)	tr (0 - 4.4)
20:5	tr (0 - 0.1)	nd ⁴	tr (0 - 0.3)
22:5	tr (0 - 0.2)	nd	nd
	Mule deer (8)	Antelope (6)	Bighorn sheep (7)
Fatty Acid			
14:0	2.3 ± 1.4 (0.4- 4.4)	3.2 ± 1.5 (1.1- 4.9)	3.6 ± 0.8 (2.0- 4.6)
14:1	tr (0.1- 1.0)	tr (0.4- 1.8)	tr (0.4- 1.3)
16:0	24.4 ± 5.7 (16.8-33.6)	19.8 ± 3.6 (15.9-26.5)	22.6 ± 1.4 (20.1-24.5)
16:1	2.2 ± 0.9 (1.0- 3.9)	2.8 ± 0.3 (2.5- 3.3)	3.5 ± 0.7 (2.4- 4.7)
18:0	26.2 ± 5.3 (18.5-34.7)	25.1 ± 3.1 (22.1-29.1)	17.2 ± 1.7 (15.7-20.2)
18:1	36.7 ± 2.6 (32.7-39.8)	36.7 ± 5.3 (28.4-45.1)	43.7 ± 1.2 (41.9-45.8)
18:2	2.7 ± 0.8 (1.2- 3.9)	3.3 ± 1.0 (2.6- 5.2)	3.0 ± 1.2 (2.2- 5.8)
18:3	1.4 ± 0.9 (0.6- 2.9)	tr (0.4- 1.3)	1.3 ± 0.2 (1.0- 1.6)
20:3	nd	tr (0 - 0.3)	nd
20:4	tr (0 - 0.3)	tr (0.2- 0.6)	tr (0.1- 0.2)
20:5	nd	tr (0 - 0.2)	tr (0 - 0.1)
22:5	tr (0 - 0.1)	nd	nd

1()= number of samples
2 = mean, standard deviation, and range
3 tr= less than 1.0%
4 nd= not detected

Table 2. Relative amounts of the major phospholipid fatty acids of longissimus dorsi muscle from some Alberta wild ruminants.

Fatty Acid	Elk (16) ¹			Moose (17)		White-tailed deer (13)	
	tr ³	(0.2- 1.1)	(0 - 0.6)	tr	(0.1- 2.6)	tr	(0.1- 0.3)
14:0	tr	(0 - 0.6)		tr	(0 - 0.7)	tr	(0 - 0.2)
14:1							
16:0	16.0 ± 6.2	(8.6-26.9)	²	16.3 ± 4.0	(10.4-24.3)	16.1 ± 2.0	(12.0-20.1)
16:1	3.5 ± 1.6	(1.1- 6.2)		1.8 ± 0.6	(0.9- 3.0)	2.0 ± 0.7	(1.2- 3.2)
18:0	15.6 ± 2.8	(12.7-23.9)		16.4 ± 2.9	(13.4-22.6)	17.0 ± 1.3	(15.3-20.1)
18:1	13.1 ± 1.8	(9.8-16.1)		15.8 ± 2.9	(10.2-21.9)	14.4 ± 3.6	(8.6-20.8)
18:2	28.1 ± 4.6	(20.7-36.2)		29.7 ± 5.6	(17.3-39.0)	29.5 ± 4.0	(22.4-34.6)
18:3	2.7 ± 0.8	(1.2- 4.0)		2.7 ± 0.9	(0.8- 4.2)	4.6 ± 1.0	(3.1- 6.5)
20:3	1.3 ± 0.4	(0.4- 2.2)		tr	(0.4- 1.2)	tr	(0.4- 1.1)
20:4	10.6 ± 2.7	(6.4-17.1)		8.6 ± 2.5	(4.4-12.4)	8.1 ± 1.5	(6.4-11.5)
20:5	2.6 ± 1.1	(1.2- 5.7)		1.8 ± 0.6	(0.9- 3.0)	2.6 ± 1.2	(1.6- 5.7)
22:5	2.4 ± 1.0	(0.8- 3.8)		1.5 ± 0.7	(0.5- 2.7)	2.4 ± 1.0	(1.3- 5.1)
Fatty Acid	Mule deer (7)			Antelope (6)		Bighorn sheep (7)	
	tr	(0.1- 1.5)	(0 - 0.6)	tr	(0.2- 1.6)	1.5 ± 1.1	(0.1- 3.0)
14:0	tr	(0 - 0.6)		tr	(0 - 0.4)	tr	(0 - 1.8)
14:1							
16:0	13.5 ± 3.5	(8.0-17.3)		16.4 ± 1.0	(15.0-17.8)	12.7 ± 1.5	(10.5-15.3)
16:1	2.1 ± 0.5	(1.2- 2.9)		1.5 ± 0.4	(1.0- 2.1)	2.6 ± 0.5	(2.0- 3.5)
18:0	18.7 ± 2.3	(15.3-21.6)		14.0 ± 1.1	(12.4-15.0)	11.2 ± 1.0	(9.9-12.8)
18:1	17.6 ± 3.6	(12.7-24.5)		13.2 ± 3.1	(9.9-18.0)	19.6 ± 2.9	(15.4-24.1)
18:2	24.0 ± 3.4	(21.1-30.6)		26.6 ± 3.2	(22.1-29.2)	21.4 ± 3.2	(16.9-26.5)
18:3	5.4 ± 1.0	(3.4- 6.5)		3.9 ± 0.4	(3.3- 4.5)	7.1 ± 0.8	(5.7- 8.1)
20:3	tr	(0.6- 1.2)		tr	(0.6- 1.1)	tr	(0.4- 0.6)
20:4	7.5 ± 2.1	(4.5-10.3)		12.5 ± 1.6	(10.4-15.0)	6.9 ± 1.1	(5.6- 9.0)
20:5	3.3 ± 0.8	(1.9- 4.4)		2.5 ± 0.7	(1.8- 3.7)	3.8 ± 0.5	(3.3- 4.9)
22:5	2.7 ± 0.6	(1.8- 3.6)		2.5 ± 0.7	(1.9- 3.6)	2.7 ± 0.4	(2.3- 3.6)

1() = number of samples
2 = mean, standard deviation and range
3 tr = less than 1.0%

1975) and the values obtained are similar to those for moose in the present study. Examination of the TG fatty acid patterns revealed that the only species that was apparently distinct from the others was elk. Elk muscle TG had higher concentrations of myristic (14:0), myristoleic (14:1), palmitic (16:0), and palmitoleic (16:1) and lower concentrations of stearic (18:0) and oleic (18:1) fatty acids. Earlier publications by Garton and Duncan (1971) and Garton et al. (1971) reported elk (and red deer) adipose tissue fatty acid patterns also differed in a similar manner from those observed in moose, white-tailed deer, reindeer and caribou adipose tissue. Concentrations of muscle TG fatty acids from the other five species examined in the present study were similar to the values obtained by Garton et al. (1971) for adipose tissue of moose and white-tailed deer except the relative concentrations of 18:0 tended to be lower and 18:1 tended to be higher. In a study of the fatty acid composition of organ fats of Lapland moose Tanhuanpää and Pulliainen (1975) obtained results for skeletal muscle similar to those obtained for this species in the present study and for perinephric and subcutaneous fat, results similar to those obtained by Garton et al. (1971). The concentrations of three fatty acids in white-tailed deer adipose tissue reported by Brüggeman et al. (1975) were within the range of values for adipose tissue from this species reported by Garton et al. (1971) and were similar to the values obtained for white-tailed deer muscle TG in the present study except the relative concentration of 18:1 was lower in muscle. Booren et al. (1973) found more 18:0 and less 18:1 in antelope kidney fat than was found in the present study for antelope l. dorsi TG. Values for the other fatty acids in antelope were similar.

The differences between species for fatty acid concentrations as

revealed by discriminant analysis are shown in Tables 3 to 6. For both TG and PL the F values for the pairwise comparison of species are all highly significant ($P < .01$) except for the F value for TG of mule deer versus white-tailed deer which was significant at $P < .05$. Wilks' lambda values of 0.011 and 0.006 and canonical correlations for the first variate of 0.953 and 0.940 were obtained for the TG and PL comparisons respectively. The first canonical variate accounted for 76% of the variation for TG but for only 59% for PL. The second variate accounted for a further 11% for TG and 24% for PL.

All twelve of the major fatty acids were useful in the discrimination using TG samples and all except 14:0 were useful for the analysis of PL. The standardized coefficients for the canonical variates in the analysis of TG show that 14:0, 14:1 and 16:1 were the main contributors to the first variate and 14:0, 14:1, 16:0 and 18:0 were the main contributors to the second variate. For the analysis using PL the main contributors to the first variate were 16:1, linoleic (18:2), linolenic (18:3) and eicosatrienoic (20:3) acids. Contributions to the second variate were fairly well spread out with 16:1 being the largest single contributor.

The distribution of the TG samples according to their scores for the first two canonical variates is shown in Figure 1. Elk are separate from the other species along the first variate but the other species all overlap with one another. Along the second variate all species overlap with the exception that bighorn sheep do not overlap with white-tailed or mule deer.

The distribution of the PL samples according to their first and second canonical variate scores is shown in Figure 2. Along the first

Table 3. Pairwise comparison of some Alberta wild ruminants using fatty acid patterns of longissimus dorsi muscle (F statistic).

<u>Triacylglycerols</u>					
Degrees of Freedom: 12, 66					
	Elk	Moose	WTD	MD	Ant
Moose	41.256				
WTD	38.295	3.367			
MD	22.287	4.028	2.138*		
Ant	19.486	4.285	5.129	6.104	
BHS	19.541	4.457	7.271	4.454	4.186

* significant at $P < .05$, all others are significant at $P < .01$

<u>Phospholipids*</u>					
Degrees of Freedom: 11, 51					
	Elk	Moose	WTD	MD	Ant
Moose	15.377				
WTD	16.104	4.835			
MD	16.004	8.577	2.639		
Ant	13.976	4.181	4.847	8.272	
BHS	30.114	17.374	11.284	6.439	13.552

*all species pairs are significantly different at $P < .01$.

Table 4. Relative importance of the canonical variates used to discriminate the longissimus dorsi muscle fatty acid patterns of some Alberta wild ruminants.

<u>Triacylglycerols</u>			Canonical Correlation
Variate	Eigenvalue	Rel. %	
1	9.888	75.90	0.953
2	1.504	11.54	0.775
3	0.956	7.34	0.699
4	0.557	4.27	0.598
5	0.123	0.95	0.331

<u>Phospholipids</u>			Canonical Correlation
Variate	Eigenvalue	Rel. %	
1	7.651	58.79	0.940
2	3.077	23.65	0.869
3	1.289	9.90	0.750
4	0.768	5.91	0.659
5	0.228	1.75	0.431

Table 5. Significance of discriminating information remaining following the derivation of successive variates.

Triacylglycerols

Variate Removed	Wilks' Lambda	Significance
0	0.011	0.000
1	0.117	0.000
2	0.292	0.000
3	0.572	0.002
4	0.890	0.388

Phospholipids

Variate Removed	Wilks' Lambda	Significance
0	0.006	0.000
1	0.049	0.000
2	0.201	0.000
3	0.460	0.000
4	0.814	0.107

Table 6. Relative contributions (standardized coefficients) of fatty acids to the canonical variates used to discriminate the fatty acid patterns of the longissimus dorsi muscle of some Alberta wild ruminants.*

		<u>Triacylglycerols</u>			
		Variate			
		1	2	3	4
Fatty Acid	14:0	0.582	1.776	-1.653	1.088
	14:1	-0.562	-1.372	1.566	-0.371
	16:0	-0.199	-2.136	-0.022	0.243
	16:1	0.840	-0.134	-0.801	0.313
	18:0	-0.063	-1.482	-0.176	0.967
	18:1	-0.317	-0.573	-0.636	0.269
	18:2	0.127	0.217	0.235	-0.672
	18:3	-0.130	-0.232	-0.694	0.428
	20:3	-0.081	-0.035	0.452	0.221
	20:4	-0.067	-0.341	-0.320	0.383
	20:5	-0.073	0.240	0.353	0.358
	22:5	0.065	-0.0145	-0.091	0.171

		<u>Phospholipids</u>			
		Variate			
		1	2	3	4
Fatty Acid	14:1	0.137	0.167	-0.327	0.451
	16:0	0.098	-0.133	-0.635	0.863
	16:1	0.392	0.814	0.052	-0.230
	18:0	0.120	0.178	-1.282	0.082
	18:1	-0.017	0.291	-0.443	-0.144
	18:2	0.346	0.358	-0.704	-0.026
	18:3	-0.516	0.414	-0.958	0.939
	20:3	0.300	0.543	-0.242	0.157
	20:4	0.161	-0.343	-0.505	0.920
	20:5	0.034	0.638	0.211	-0.246
	22:5	0.084	-0.401	-0.463	0.308

* Variate 5 is not shown because of its low significance.

Figure 1. Distribution of longissimus dorsi triacylglycerol samples by species in two-dimensional discriminant space.

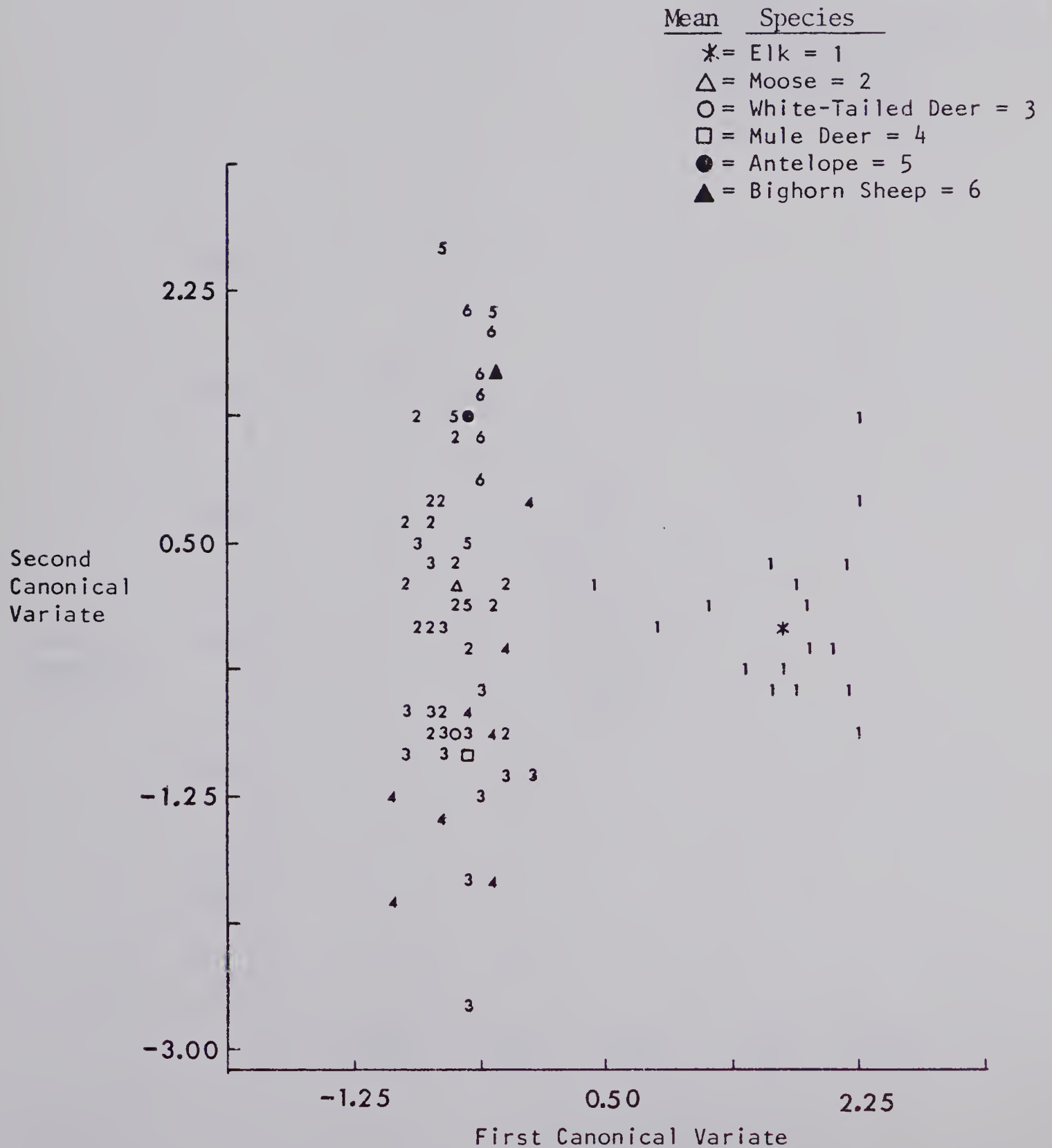
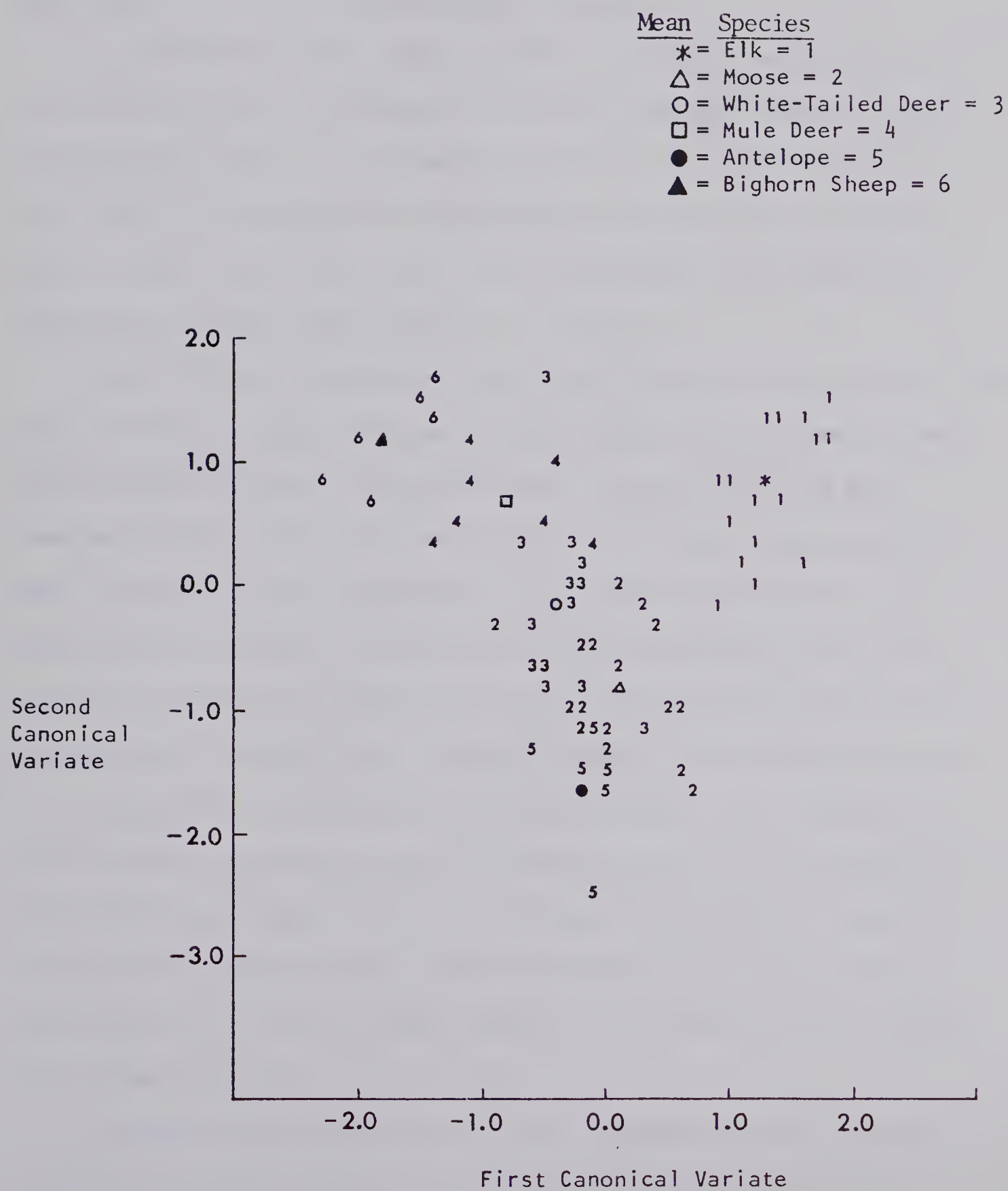


Figure 2. Distribution of longissimus dorsi phospholipid samples by species in two-dimensional discriminant space.



variate elk do not overlap with the other species, bighorn sheep only overlap with mule deer, and the remaining four species all overlap each other. Along the second variate antelope do not overlap with elk, mule deer or bighorn sheep and moose do not overlap with mule deer or bighorn sheep but all other species pairs overlap.

As indicated by the F values in Table 3 the fatty acid patterns of elk muscle were quite different from those of the other species. Moose and white-tailed deer were somewhat similar and, as might be expected on the basis of phylogenetic relationships, white-tailed deer and mule deer were the most similar pair. The relationship of the species to one another is more readily apparent in Figure 1.

From the results obtained in the present study and from the published work of Garton and his colleagues it would appear that elk and red deer have an unique TG fatty acid pattern among ruminants that have been examined to date. These two types of deer are closely related and have been classified as the same species. A possible explanation for the observed unique pattern is that during their evolutionary history the ancestors of these two members of the deer family may have experienced a mutation(s) affecting their synthesis of fatty acids whereby they had a reduced ability to lengthen fatty acids following their release from the fatty acid synthetase complex. This would result in an accumulation of straight chain fatty acids 14 and 16 carbon atoms long. It would be of interest to determine the TG fatty acid pattern in other members of this genus and in closely related species to determine how far reaching this unique pattern is.

Although there appeared to be little difference between species in PL fatty acid pattern (Table 2) the species differences were again

highly significant as indicated by the discriminant analysis. Again, elk were quite different from the other species and white-tailed deer and mule deer were the most similar. A comparison of the distribution of PL samples (Figure 2) to that for TG samples (Figure 1) shows that the overall separation of the species studied is much clearer using PL samples than using the TG samples.

Tanhuanpää and Pulliainen (1975) pointed out that the results of Garton and Duncan (1971) and Garton et al. (1971) showed a greater similarity in fatty acid composition between closely related species (i.e. red deer and elk, caribou and reindeer) than between species not closely related that were consuming similar diets (i.e. moose and elk). Examination of the fatty acid composition of abomasal contents from several elk, moose and white-tailed deer during the present study showed that there were no notable species differences (Table 7). This observation supports the hypothesis that the diet of these species has a minimal effect on the observed differences in the fatty acid composition of their tissues. This hypothesis is further supported by the fact that adipose tissues of elk from Alberta consuming mainly browse (Garton et al., 1971) and of the closely related red deer from Scotland consuming heather (Garton and Duncan, 1971) have similar fatty acid patterns as do the adipose tissues of moose from Alberta consuming mainly browse from deciduous trees (Garton et al., 1971) and moose from Lapland consuming mainly leaves and needles of pine and juniper (Tanhuanpää and Pulliainen, 1975). In addition, although the samples for some species in the present study came from different habitats and at various times of the year the fatty acid patterns did not appear to be affected. Similarly, Tanhuanpää and Pulliainen (1975) have reported seasonal diet

Table 7. Relative concentrations of major fatty acids in the abomasal contents from individual elk, moose and white-tailed deer.

Fatty Acid	Elk ¹						Moose			White-Tailed Deer	
	1	2	3	4	5	6	1	2	3	1	2
14:0	5.8	1.6	1.9	0.6	1.4	1.4	0.7	1.6	1.5	1.8	2.6
16:0	19.4	13.1	15.7	13.7	12.1	14.2	9.9	8.6	16.0	8.3	10.6
16:1	2.9	3.5	1.7	2.2	1.2	2.0	2.4	0.9	1.8	1.5	1.1
17:0	1.7	2.4	1.9	1.8	1.2	2.2	1.3	0.9	2.5	0.9	1.2
18:0	23.9	33.4	30.8	29.1	42.2	45.6	38.1	37.4	36.3	43.2	33.3
18:1	15.5	13.7	12.4	17.6	12.2	12.8	14.2	12.4	10.8	11.2	9.6
18:2	5.5	5.2	6.9	12.1	6.4	4.9	10.6	8.5	7.4	7.4	7.0
18:3	1.4	1.1	2.8	1.3	3.9	2.1	2.4	1.9	3.6	2.0	1.6
20:0	2.2	3.6	2.3	3.4	2.2	2.2	4.4	3.8	2.9	3.5	6.4
22:0	1.9	3.0	2.0	2.0	2.0	1.7	9.0	3.0	2.7	2.7	3.1
U ²	1.7	2.9	2.9	2.3	1.8	2.1	7.4	4.4	2.6	2.5	4.1
U	4.7	8.3	2.0	2.2	2.0	4.0	1.4	4.4	2.0	1.4	1.9

¹ Elk: 1=calf, Dec.28/76; 2=adult female, Dec.28/76; 3=5½ year male, Jan.27/77; 4=yearling male, Mar.9/77; 5=adult female, May 18/77; 6=adult female, Aug.24/77.
Moose: 1=female calf, Jan.4/77; 2=3½ year male, Jan.25/77; 3=1 year female, May 18/77.
White-Tailed Deer: 1=yearling male, Jan.23/77; 2=female (yearling or adult), Feb.1/77.
² U = Unidentified fatty acid.

changes appeared to have little effect on tissue fatty acid composition. Tove (1965) indicates that changes in the fatty acid composition of the diet have a negligible effect on fatty acid composition of ruminant fats when normal forage diets are fed. However, special diets such as finishing rations have been found to affect tissue fatty acid composition (Rumsey et al., 1972; Sumida et al., 1972; Duncan et al., 1974; Shaw et al., 1960). During the past few years the use of the technique of feeding protected lipids to ruminants has made it possible to exert a great influence on the fatty acid composition of their tissues. Feeding protected lipids and finishing rations are situations not encountered by big game animals.

Changes in dietary conditions encountered by wild ruminants may result in an altered rumen flora which could influence the hydrogenation of unsaturated fatty acids and the proportions of volatile fatty acids produced thereby influencing tissue fatty acid composition (Brüggeman et al., 1975; Shaw et al., 1960). Roberts (1966) found that while the proportions of rumen long-chain fatty acids showed marked differences due to diet there was little correlation of these differences with the proportions of the major long chain fatty acids deposited in the carcass. Roberts (1966) suggested that the effect of diet on the relative amounts of volatile fatty acids in the rumen may have resulted in an altered endogenous fatty acid synthesis and thereby resulted in the observed differences in tissue fatty acid composition. However, the differences he observed for intramuscular fat from animals on hay versus grain diets, although sometimes significant at $P < .01$, were usually small. Short et al. (1966), in a study on rumen volatile fatty acid levels in free-ranging deer, found that during a one year period acetic

acid varied between a high of 70% and low of 60%, proprionic stayed about 20 to 23% and butyric varied between 9 and 14%. If such degrees of variation are typical for wild ruminants this factor would appear to have little possibility for significantly affecting endogenous fatty acid synthesis.

In summary, significant differences among all species for both TG and PL fatty acid patterns were obtained. Elk were the most distinct while mule deer and white-tailed deer were the least distinct. Diet and time of year would appear to have little effect on differences in fatty acid patterns observed in the present study.

CHAPTER 2 Age and sex related differences in triacylglycerol and phospholipid fatty acid patterns from l. dorsi muscle of elk, moose and white-tailed deer

INTRODUCTION

It is well recognized that extensive changes occur in the metabolism of ruminant species between the neonatal stage and the time when the rumen becomes fully functional. The effect of this transition on the fatty acid composition of various tissues from domestic ruminants has been well documented (Hecker et al., 1975; Leat, 1970). Changes in tissue fatty acid composition with age after the rumen has become functional have also been reported (Clemens et al., 1973; Hecker et al., 1975).

There has been a considerable amount of information published describing the effect of sex on the fatty acid composition of tissues from domestic animals. However the only reports dealing with the effect of sex on tissue fatty acid composition of wild ruminants only compare one animal of each sex in the case of elk, moose, white-tailed deer and caribou (Garton et al., 1971) and one male versus two female reindeer (Garton and Duncan, 1971).

Chapter 2 will provide an examination of the fatty acid pattern of the l. dorsi as a function of age and as function of sex in elk, moose and white-tailed deer.

EXPERIMENTAL

The preparation and chemical analysis of muscle samples are outlined in Chapter 1. Comparisons on the basis of age and sex were only made for elk, moose and white-tailed deer as there were insufficient

samples of the other species referred to in this study.

In the analysis of the relative amounts of the fatty acids by animal age, comparisons for both elk and moose were made between samples from fetuses, from animals seven to nine months of age and from animals greater than fifteen months of age. For white-tailed deer, samples from animals four to fifteen months of age were compared to samples from animals over fifteen months of age. There were no fetal samples available for white-tailed deer nor were there samples from elk or moose four to six months of age.

Fetal samples were not included when comparing samples by sex.

RESULTS AND DISCUSSION

Age

The relative concentrations of the major fatty acids of the l. dorsi TG and PL from elk grouped by age are shown in Table 8, from moose in Table 9 and from white-tailed deer in Table 10. Fatty acid values for individual animals are tabulated in Appendix 1.

Examination of the fatty acid concentrations reveals that there was no consistent fatty acid pattern among the three species. However, there appear to be some trends within species and between species for some individual fatty acids.

For elk the TG fetal samples were more different overall from the seven to nine month group than from the oldest age group. The fetal samples had lower values for 14:1 than the oldest age group but the two groups overlap for values of the other fatty acids. The fetal samples

Table 8. Relative amounts of the major fatty acids of longissimus dorsi muscle from elk of different ages.

Fatty Acid	Triacylglycerols		
	Fetus (2) ¹	7 to 15 months (2)	>15 months (16) ²
14:0	3.5 (3.1- 3.9)	4.3 (2.8- 5.8)	6.9 ± 2.1 (3.5-11.6)
14:1	tr ³ (0.1- 0.5)	tr (0.3- 0.6)	3.0 ± 1.0 (1.4- 5.0)
16:0	42.6 (39.9-45.4)	33.0 (25.9-40.0)	35.7 ± 4.6 (27.8-43.7)
16:1	6.3 (5.8- 6.8)	3.4 (2.7- 4.0)	15.6 ± 3.5 (6.0-20.6)
18:0	15.6 (13.6-17.7)	23.4 (19.2-27.6)	8.7 ± 4.1 (5.4-21.9)
18:1	23.2 (22.5-23.8)	24.4 (21.4-27.5)	23.6 ± 3.3 (17.6-29.0)
18:2	2.6 (2.5- 2.7)	4.8 (4.0- 5.5)	2.7 ± 1.4 (1.2- 5.9)
18:3	tr (0.1- 0.3)	tr ⁴ (0.4)	tr (0.1- 1.2)
20:3	tr (0 - 0.4)	nd	tr (0 - 0.1)
20:4	1.8 (1.6- 1.9)	nd	tr (0 - 1.1)
20:5	nd	nd	tr (0 - 0.1)
22:5	tr (0 - 0.6)	nd	tr (0 - 0.2)
Fatty Acid	Phospholipids		
	Fetus (1)	7 to 15 months (2)	>15 months (14)
14:0	tr	tr (0.3- 0.5)	tr (0.2- 1.1)
14:1	nd	tr (0 - 0.1)	tr (0 - 0.6)
16:0	28.4	10.6 (8.6-12.7)	17.2 ± 6.2 (9.2-30.2)
16:1	2.7	1.3 (1.1- 1.5)	3.9 ± 1.4 (2.1- 6.2)
18:0	19.8	15.4 (14.7-16.0)	15.6 ± 3.1 (12.7-23.9)
18:1	29.2	10.0 (9.8-10.2)	13.6 ± 1.4 (11.7-16.1)
18:2	2.4	35.9 (35.6-36.2)	27.3 ± 3.8 (20.7-31.8)
18:3	tr	2.8 (2.6- 3.1)	2.8 ± 0.8 (1.2- 4.0)
20:3	tr	1.5 (1.4- 1.6)	1.3 ± 0.4 (0.4- 2.2)
20:4	7.0	11.9 (10.4-13.4)	10.6 ± 2.9 (6.4-17.1)
20:5	tr	4.8 (3.9- 5.7)	2.4 ± 0.8 (1.2- 3.9)
22:5	2.7	3.3 (2.8- 3.8)	2.5 ± 0.8 (0.8- 3.7)

1()=number of samples; 2 = mean, standard deviation, and range; 3 tr = less than 1.0%; 4 nd = not detected

Table 9. Relative amounts of the major fatty acids of longissimus dorsi from moose of different ages.

Triacylglycerols				
Fatty Acid	Fetus (5) ¹		7 to 15 months (3)	
	3.2 ± 0.4 (2.6- 3.8) ²	tr ³ (0 - 0.2)	2.2 ± 0.4 (1.9- 2.6)	1.0 ± 0.4 (0.5- 2.0)
14:0			tr	tr (0 - 0.8)
14:1				
16:0	38.9 ± 1.5 (37.0-40.4)		21.7 ± 1.6 (20.3-23.5)	19.1 ± 2.5 (15.5-23.2)
16:1	9.3 ± 0.6 (8.5-10.1)		2.7 ± 0.1 (2.6- 2.8)	2.4 ± 1.0 (1.5- 5.0)
18:0	14.0 ± 0.8 (13.3-15.2)		25.7 ± 1.3 (24.9-27.2)	24.0 ± 4.8 (11.7-33.6)
18:1	27.1 ± 1.2 (25.4-28.3)		33.8 ± 3.2 (31.0-37.3)	44.2 ± 4.6 (34.5-56.2)
18:2	2.1 ± 0.3 (1.6- 2.4)		4.4 ± 1.0 (3.4- 5.4)	3.5 ± 0.8 (2.0- 5.2)
18:3	tr (0.1- 0.8)		1.3 ± 0.5 (0.7- 1.6)	tr (0.3- 1.7)
20:3	tr (0.5- 1.0)		tr (0 - 0.1)	tr (0 - 0.1)
20:4	tr (0.4- 0.9)		tr (0 - 0.3)	tr (0 - 0.3)
20:5	tr ⁴ (0 - 0.2)		nd	nd
22:5	nd		nd	nd
Phospholipids				
Fatty Acid	Fetus (5)		7 to 15 months (3)	
	tr (0.7- 1.0)	nd	tr (0.1- 0.6)	tr (0.1- 2.6)
14:0				tr (0 - 0.7)
14:1				
16:0	24.8 ± 1.7 (23.2-27.4)		16.5 ± 4.6 (11.8-20.9)	16.3 ± 4.0 (10.4-24.3)
16:1	3.0 ± 0.4 (2.5- 3.5)		1.6 ± 0.4 (1.3- 2.1)	1.8 ± 0.6 (0.9- 3.0)
18:0	19.8 ± 1.3 (18.7-21.9)		16.4 ± 2.7 (13.9-19.3)	16.4 ± 3.0 (13.4-22.6)
18:1	31.5 ± 2.4 (28.2-34.8)		15.0 ± 2.5 (12.1-16.5)	15.9 ± 3.1 (10.2-21.9)
18:2	1.9 ± 0.5 (1.3- 2.5)		29.9 ± 3.4 (26.0-32.0)	29.7 ± 6.0 (17.3-39.0)
18:3	tr (0.1- 0.2)		2.9 ± 0.6 (2.3- 3.4)	2.7 ± 1.0 (0.8- 4.2)
20:3	1.2 ± 0.2 (0.9- 1.5)		tr (0.4- 1.0)	tr (0.4- 1.2)
20:4	6.2 ± 0.7 (5.1- 7.0)		7.8 ± 3.2 (5.4-11.5)	8.7 ± 2.4 (4.4-12.4)
20:5	tr (0.3- 0.9)		1.8 ± 1.1 (0.9- 3.0)	1.8 ± 0.6 (1.0- 2.9)
22:5	2.7 ± 1.0 (1.5- 4.2)		1.3 ± 0.8 (0.5- 2.1)	1.5 ± 0.7 (0.6- 2.7)

1 () = number of samples; 2 = mean, standard deviation, and range; 3 tr=less than 1.0%; 4 nd=not detected

Table 10. Relative amounts of the major fatty acids of longissimus dorsi from white-tailed deer of different ages.

		<u>Triacylglycerols</u>	
		4 to 15 months (10) ¹	>15 months (10)
Fatty Acid	14:0	1.7 ± 0.7 (0.7- 2.7) ²	1.0 ± 0.4 (0.3- 1.5)
	14:1	tr ³ (0.2- 0.6)	tr (0 - 0.5)
	16:0	23.7 ± 3.9 (17.6-30.6)	20.5 ± 2.2 (17.9-24.8)
	16:1	2.7 ± 0.8 (1.7- 4.1)	2.0 ± 0.7 (1.4- 3.8)
	18:0	27.9 ± 5.9 (19.8-40.8)	28.7 ± 3.7 (22.6-34.5)
	18:1	34.6 ± 5.0 (24.4-40.5)	40.0 ± 6.4 (27.6-48.9)
	18:2	3.3 ± 0.8 (2.3- 5.1)	3.3 ± 2.2 (1.9- 9.4)
	18:3	1.2 ± 0.5 (0.4- 1.8)	tr (0.2- 1.4)
	20:3	tr (0 - 0.1)	tr (0 - 0.1)
	20:4	tr (0 - 0.6)	tr (0 - 4.4)
	20:5	tr ⁴ (0 - 0.3)	tr (0 - 0.2)
	22:5	nd	nd

		<u>Phospholipids</u>	
		4 to 15 months (5)	>15 months (8)
Fatty Acid	14:0	tr (0.1- 0.2)	tr (0.1- 0.3)
	14:1	tr (0 - 0.1)	tr (0 - 0.2)
	16:0	14.4 ± 1.5 (12.0-15.8)	17.1 ± 1.7 (15.6-20.1)
	16:1	1.9 ± 0.8 (1.2- 3.2)	2.0 ± 0.6 (1.4- 3.1)
	18:0	16.9 ± 0.8 (15.8-18.0)	17.0 ± 1.5 (15.3-20.1)
	18:1	13.5 ± 4.5 (8.6-20.8)	15.0 ± 3.1 (10.1-18.4)
	18:2	29.0 ± 5.9 (22.4-34.6)	29.9 ± 2.8 (25.8-34.0)
	18:3	5.3 ± 0.9 (4.5- 6.5)	4.2 ± 0.8 (3.1- 5.4)
	20:3	tr (0.7- 1.1)	tr (0.4- 0.6)
	20:4	8.2 ± 2.0 (6.4-11.5)	8.0 ± 1.3 (6.8-10.2)
	20:5	3.7 ± 1.4 (2.4- 5.7)	2.0 ± 0.4 (1.6- 2.6)
	22:5	3.2 ± 1.2 (2.2- 5.1)	1.8 ± 0.4 (1.3- 2.3)

1 () = number of samples

2 = mean, standard deviation, and range

3 tr = less than 1.0%

4 nd = not detected

had more 16:0 and 16:1, less 18:0 and 18:2 and similar values for the other fatty acids when compared to the seven to nine month group.

The single PL fetal sample was quite different from the samples in the other two age groups. It had more 18:1 and less 18:2, 18:3 and 20:5 than either of the other two groups. For the values of the other fatty acids it overlapped with the samples from the oldest age group but it had more 16:0, 16:1 and 18:0 and less 20:3 and 22:5 than the samples in the seven to nine month group.

For TG of moose the fetal samples were much different from those in the other two age groups. They had higher 16:0 and 16:1 and lower 18:1 than the older animals. The fetal samples overlapped with the oldest age group for the other fatty acids except 14:0 for which they had slightly higher values. The fetal samples had similar values to the seven to nine month group for 14:0 and 14:1 but they had less 18:0, 18:2 and 18:3. For PL the fetal samples were again quite different from the older two age groups. The fetal samples had more 18:1 and less 18:2 and 18:3. Compared to the seven to nine month group the fetal samples had overlapping values for the other fatty acids except that they had more 16:0 and 16:1. The fetal samples overlapped with the oldest age group for all of the fatty acid values except 18:1, 18:2 and 18:3 as mentioned above.

A comparison of the elk and moose fetal samples indicates that in general the TG and PL fatty acid patterns were similar. However, elk had relatively lower amounts of TG 16:1 and 18:1 than moose. For both elk and moose samples TG 16:0 and 16:1 decreased and 18:0 and 18:2 increased in going from the fetal stage to the age of seven to nine months. This same comparison for PL showed that for both species 16:0,

16:1, 18:0 and 18:1 values decreased and 18:2, 18:3, 20:4 and 20:5 increased.

The only published data on the muscle fatty acid composition of very young wild ruminants that was found was from work done by Payne. The present study supports the findings of Payne (1978) who reported that muscle of fetal and newborn ruminants had relatively less PL 18:2 and 18:3 and more 18:1 than mature ruminants. Also in agreement with Payne (1978) there was little difference in PL 20:4 levels between fetal and adult samples in the present study. Payne (1978) reported that in his study neonatal red deer had more PL 18:2 than fetal calves or neonatal lambs. However the PL 18:2 values for fetal elk, and also moose, in the present study are more similar to Payne's calves and lambs than to his red deer calves. The other PL fatty acid values of the single elk fetus in the present study also differed from the mean values for the two fetuses reported by Payne (1978) but it would be necessary to increase the sample sizes to determine if there are significant differences between elk and red deer fetuses. The low levels of 18:2 and 18:3 in the samples from fetal elk and moose may indicate that these animals are near an essential fatty acid deficient state. Payne (1978) suggested that because the relative amount of 20:4 is fairly constant in animals of all ages it may be the really essential fatty acid with 18:2 acting as a store available for conversion to 20:4 as needed.

For elk the TG 14:1 and 16:1 values are higher and the 18:0 values tend to be lower in the greater than fifteen month age group than in the seven to fifteen month group. Contrary to this, Clemens et al. (1973) found no age related trends in the fatty acid composition of ether

extracts of bovine l. dorsi muscle. For PL the seven to nine month group differed from the older age group in that they had less 16:1 and 18:1 and more 18:2 and 20:5. Hecker et al. (1975) reported an increase in the degree of unsaturation of bovine muscle total fatty acids as the animals grew from 28 days of age to 1.5 years of age. They ascribed this mainly to an increase in the amount of 18:1 and a decrease in 18:0. Other changes noted were an increase in 14:1 and 16:1 and a decrease in 18:2. The authors felt that although changes in the proportions of the various lipid classes may have had some effect on the changing fatty acid pattern it was likely that the desaturase system was becoming more active with growth. The results for both TG and PL fatty acids of elk tend to agree with the patterns seen by Hecker et al. (1975). The sample size for elk in the seven to fifteen month group was very small and the initial agreement with the results of Hecker et al. (1975) might easily disappear with a larger sample size.

For moose TG the 14:0 values are lower and the 18:1 values tend to be higher in samples from the oldest age group. There was little difference between the older two age groups in PL fatty acid values. These results do not agree with those of either Clemens et al. (1973) or Hecker et al. (1975).

There was little difference in TG fatty acid values between the two age groups for white-tailed deer which would agree with the results of Clemens et al. (1973). For PL the samples from animals less than fifteen months old tended to have less 16:0 and more 18:3, 20:3, 20:5 and 22:5. These fatty acids are not the same as those that were found to vary with animal age by Hecker et al. (1975). Within the younger age

group the sample from the animal in the four to six month bracket had the highest value for 16:0, 16:1, 18:3, 20:5 and 22:5. Contrary to this Hecker et al. (1975) reported 16:1 to increase with animal age. The sample from the animal in the ten to twelve month bracket had the lowest value for 16:0 and 18:1 and the highest value for 20:3 and 20:4 and the samples from the seven to nine month bracket had the highest values for 18:1 and the lowest values for 18:3, 20:5 and 22:5. Unlike the results of Hecker et al. (1975) the 18:1 values here did not show a consistent change with age. It is apparent then that none of these fatty acids showed any consistent change with increasing animal age.

Changes in fatty acid values with age for animals over three months of age showed no consistent trends among the three species. Most of the trends that were seen were of small magnitude and could easily disappear with larger sample sizes. However, an interesting pattern was observed for TG when 18:1 as a percent of the sum of 14:1, 16:1 and 18:1 was plotted for known age animals (Figure 3). The values for moose and elk fetuses overlap and the value was greater in calves of both species and similar to values for white-tailed deer fawns. The values for older age moose and deer were similar to the values for calves and fawns. For elk, however, the values for yearling and older elk were much lower than the values of the calves. It would appear then that for elk at least age does influence TG fatty acid composition.

Sex

The values for the major fatty acids of l. dorsi muscle TG and PL from elk grouped by sex are shown in Table 11, for moose in Table 12, and for white-tailed deer in Table 13. Fatty acid values for individual

Table 11. Relative amounts of the major fatty acids of longissimus dorsi from male and female elk.

		<u>Triacylglycerols</u>	
		Male (6) ¹	Female (12)
Fatty Acid	14:0	6.8 ± 3.3 (2.8-11.6) ²	6.5 ± 1.5 (4.7- 9.4)
	14:1	2.1 ± 1.3 (0.3- 3.6)	3.0 ± 1.2 (0.6- 5.0)
	16:0	35.3 ± 7.5 (25.9-43.7)	35.5 ± 3.7 (29.3-41.5)
	16:1	10.6 ± 5.2 (2.7-16.9)	16.0 ± 4.2 (4.0-20.6)
	18:0	14.4 ± 8.4 (7.4-27.6)	8.3 ± 3.9 (5.4-19.2)
	18:1	22.8 ± 4.6 (17.6-29.0)	24.1 ± 2.6 (20.5-28.4)
	18:2	3.1 ± 1.6 (1.5- 5.9)	2.8 ± 1.5 (1.2- 5.9)
	18:3	tr ³ (0.1- 1.5)	tr ⁴ (0.2- 1.2)
	20:3	tr (0 - 0.1)	nd ⁴
	20:4	tr (0 - 1.1)	tr (0 - 1.0)
	20:5	tr (0 - 0.1)	nd
	22:5	tr (0 - 0.2)	nd

		<u>Phospholipids</u>	
		Male (6)	Female (10)
Fatty Acid	14:0	tr (0.2- 1.0)	tr (0.3- 1.1)
	14:1	tr (0 - 0.2)	tr (0 - 0.6)
	16:0	12.7 ± 4.6 (8.6-21.4)	18.5 ± 6.3 (9.3-30.2)
	16:1	2.5 ± 0.8 (1.1- 3.3)	4.2 ± 1.6 (1.5- 6.2)
	18:0	15.8 ± 2.6 (12.9-19.7)	15.4 ± 3.2 (12.7-23.9)
	18:1	12.9 ± 1.9 (10.2-15.4)	13.4 ± 1.8 (9.8-16.1)
	18:2	30.3 ± 3.7 (25.8-36.2)	27.2 ± 4.9 (20.7-35.6)
	18:3	3.0 ± 1.0 (1.2- 4.0)	2.6 ± 0.6 (1.7- 3.4)
	20:3	1.6 ± 0.4 (1.3- 2.2)	1.1 ± 0.3 (0.4- 1.4)
	20:4	12.5 ± 2.7 (9.3-17.1)	9.8 ± 2.4 (6.4-13.8)
	20:5	3.1 ± 1.6 (1.5- 5.7)	2.4 ± 0.8 (1.2- 3.9)
	22:5	2.8 ± 1.2 (0.8- 3.8)	2.4 ± 0.6 (1.2- 3.3)

1 () = number of samples

2 = mean, standard deviation and range

3 tr = less than 1.0%

4 nd = not detected

Table 12. Relative amounts of the major fatty acids of longissimus dorsi from male and female moose.

		<u>Triacylglycerols</u>	
		Male (11) ¹	Female (12)
Fatty Acid	14:0	1.1 ± 0.6 (0.5- 2.0) ²	1.3 ± 0.6 (0.7- 2.6)
	14:1	tr ³ (0.2- 0.8)	tr (0 - 0.8)
	16:0	18.2 ± 2.3 (15.5-21.6)	20.6 ± 2.1 (17.1-23.5)
	16:1	2.6 ± 1.2 (1.6- 5.0)	2.3 ± 0.7 (1.5- 4.1)
	18:0	23.5 ± 5.4 (11.7-29.2)	24.9 ± 3.6 (19.5-33.6)
	18:1	44.2 ± 5.4 (36.4-56.2)	41.6 ± 5.9 (31.0-48.4)
	18:2	3.5 ± 0.8 (2.1- 4.4)	3.6 ± 1.0 (2.0- 5.4)
	18:3	1.0 ± 0.4 (0.3- 1.7)	1.0 ± 0.4 (0.6- 1.6)
	20:3	tr (0 - 0.1)	tr (0 - 0.1)
	20:4	tr ⁴ (0 - 0.3)	tr (0 - 0.3)
	20:5	nd	nd
	22:5	nd	nd

		<u>Phospholipids</u>	
		Male (8)	Female (9)
Fatty Acid	14:0	tr (0.1- 2.6)	tr (0.1- 0.9)
	14:1	tr (0 - 0.7)	tr (0 - 0.5)
	16:0	14.4 ± 2.2 (10.4-16.8)	18.1 ± 4.4 (11.8-24.3)
	16:1	1.7 ± 0.6 (1.0- 2.6)	1.8 ± 0.6 (0.9- 3.0)
	18:0	14.8 ± 1.0 (13.4-16.5)	17.9 ± 3.3 (13.7-22.6)
	18:1	15.8 ± 3.7 (10.2-21.9)	15.7 ± 2.3 (12.1-19.2)
	18:2	30.8 ± 7.2 (17.3-39.0)	28.7 ± 3.9 (23.6-36.2)
	18:3	3.0 ± 0.7 (1.8- 4.2)	2.4 ± 1.0 (0.8- 4.0)
	20:3	tr (0.4- 1.2)	tr (0.4- 1.0)
	20:4	9.6 ± 2.3 (6.5-12.4)	7.6 ± 2.4 (4.4-11.5)
	20:5	1.9 ± 0.5 (1.0- 2.4)	1.7 ± 0.8 (0.9- 3.0)
	22:5	1.4 ± 0.4 (1.0- 2.2)	1.5 ± 0.9 (0.5- 2.7)

1 () = number of samples

2 = mean, standard deviation, and range

3 tr = less than 1.0%

4 nd = not detected

Table 13. Relative amounts of the major fatty acids of longissimus dorsi from male and female white-tailed deer.

		<u>Triacylglycerols</u>	
		Male (4) ¹	Female (6)
Fatty Acid	14:0	1.4 ± 0.7 (0.3- 2.7) ²	1.3 ± 0.7 (0.6- 2.7)
	14:1	tr ³ (0 - 0.6)	tr (0 - 0.5)
	16:0	22.4 ± 4.1 (18.2-30.6)	21.8 ± 3.1 (17.6-27.8)
	16:1	2.6 ± 1.0 (1.4- 3.8)	2.1 ± 0.5 (1.4- 3.1)
	18:0	28.0 ± 4.6 (19.8-34.5)	28.5 ± 5.2 (21.8-40.8)
	18:1	36.2 ± 4.8 (27.6-43.4)	38.2 ± 7.3 (24.4-48.9)
	18:2	3.6 ± 2.3 (1.9- 9.4)	3.1 ± 0.8 (2.3- 5.1)
	18:3	1.0 ± 0.5 (0.2- 1.4)	1.0 ± 0.4 (0.4- 1.8)
	20:3	tr (0 - 0.1)	tr (0 - 0.1)
	20:4	tr (0 - 4.4)	tr (0 - 0.6)
	20:5	tr ⁴ (0 - 0.2)	tr (0 - 0.3)
	22:5	nd ⁴	nd

		<u>Phospholipids</u>	
		Male (5)	Female (8)
Fatty Acid	14:0	tr (0.1- 0.3)	tr (0.1- 0.3)
	14:1	tr (0 - 0.2)	tr (0 - 0.2)
	16:0	16.8 ± 2.0 (15.4-20.1)	15.6 ± 2.1 (12.0-19.3)
	16:1	2.3 ± 0.9 (1.2- 3.2)	1.8 ± 0.5 (1.4- 2.8)
	18:0	17.4 ± 1.5 (16.2-20.1)	16.7 ± 1.1 (15.3-18.3)
	18:1	12.1 ± 1.4 (10.1-13.8)	15.9 ± 3.9 (8.6-20.8)
	18:2	28.9 ± 4.4 (23.3-34.6)	30.0 ± 4.0 (22.4-34.5)
	18:3	4.9 ± 1.2 (3.1- 6.5)	4.5 ± 0.8 (3.9- 5.9)
	20:3	tr (0.6- 0.9)	tr (0.4- 1.1)
	20:4	8.3 ± 1.7 (6.4-10.2)	8.0 ± 1.5 (6.8-11.5)
	20:5	3.0 ± 1.5 (2.0- 5.7)	2.4 ± 1.0 (1.6- 4.1)
	22:5	2.8 ± 1.3 (2.1- 5.1)	2.1 ± 0.8 (1.3- 3.8)

1 () = number of samples

2 = mean, standard deviation and range

3 tr = less than 1.0%

4 nd = not detected

samples are tabulated in Appendix 1.

In the present study the only fatty acid for which males and females had fairly distinct ranges in values was 20:3 in elk PL. Some of the other fatty acids tended to be different between males and females but in all cases there were considerable overlaps in the ranges of values for each of the fatty acids. When only animals over one year of age were considered the only additional differences between the sexes noted were that white-tailed deer TG and PL 18:1 concentrations were higher in females and PL 20:4 and 22:5 concentrations were lower in females than males. Hood and Allen (1971) found that while the fatty acid composition of intramuscular PL, TG, mono- and diglyceride and free fatty acid fractions from heifers and steers was similar, that of bulls contained relatively more TG 18:0 and PL 18:2. Neither of these fatty acids differed between sexes in the three species examined in the present study. The finding in the present study that the muscle fatty acid composition of males and females is very similar is in agreement with the results of Tanhuanpää and Pulliainen (1975) for adipose tissues of male and female moose. Garton and Duncan (1971) reported that lactation lowered the relative amount of 18:0 in depot fat of red deer hinds. If this is also true for muscle lipid it would make females in this condition more distinct from males than adult pregnant and normal females are.

In summary, there was no consistent age related change in muscle TG or PL fatty acid patterns among the three species. However, there were some trends within species and between pairs of species for some fatty acids. Fetuses were always distinct from older animals. Male and female groups for each of the three species had very similar fatty acid patterns.

CHAPTER 3 The effect of anatomical location on the fatty acid composition of muscle

INTRODUCTION

In Chapters 1 and 2 all comparisons were based on analysis of 1. dorsi. Since it has been reported that the fatty acid composition of meat can vary with anatomical location (Gillis et al., 1973; Hornstein et al., 1968; O'Keefe et al., 1968) the effect of this variable on muscle fatty acid pattern of wild ruminants was investigated.

EXPERIMENTAL

Sample preparation and analysis are outlined in Chapter 1. The effect of anatomical location on the relative proportions of individual fatty acids was determined by comparing the values for muscles from up to six different sites: 1) lower front leg (extensor capri radialis muscle), 2) shoulder (infraspinatus muscle), 3) loin (1. dorsi muscle), 4) rump (biceps femoris muscle), 5) rump (semitendinosus muscle), 6) lower hind leg (long digital extensor and peroneus tertius). Multiple muscle samples were taken from seven elk, four moose, five white-tailed deer, one antelope and one bighorn sheep.

RESULTS AND DISCUSSION

Fatty acids in the PL fraction showed little variation with anatomical location therefore only the values for the major TG fatty acid concentrations will be presented. The fatty acid composition of the TG and PL fractions of each muscle sample are tabulated in Appendix I.

Elk

Results for elk are shown in Figure 4. For all individuals, except the calf, the muscles of the extremities tended to have lower values for 16:0 and higher values for 18:1 than did the more centrally located muscles. For the calf this trend in 16:0 values was still apparent but the trend for 18:1 seen in the older animals was not present. For the two animals collected in January, the adult female collected in March and the calf there seemed to be a slight tendency for 16:1 values to be higher and 18:0 values to be lower in the muscles from the extremities than in the more central muscles. The trends for these two fatty acids are not apparent in the other individuals.

Moose

The results for the analysis of the TG fatty acids of different muscles from four moose are shown in Figure 5. Very little variation in fatty acid values occurred in the one year old female. The adult male and the female calf both showed little variation for 16:0 or 16:1 but in both animals there was a tendency for the muscles from the extremities to have lower values for 18:0 and higher values for 18:1 than did the more central muscles. The adult female showed quite a bit of variability for all four fatty acids shown. Values for 16:0 and 16:1 tended to be higher in muscles from the extremities than in the other muscles and the 18:0 and 18:1 values showed the inverse of this trend.

White-tailed deer

Data on the variability of fatty acid values between muscles from

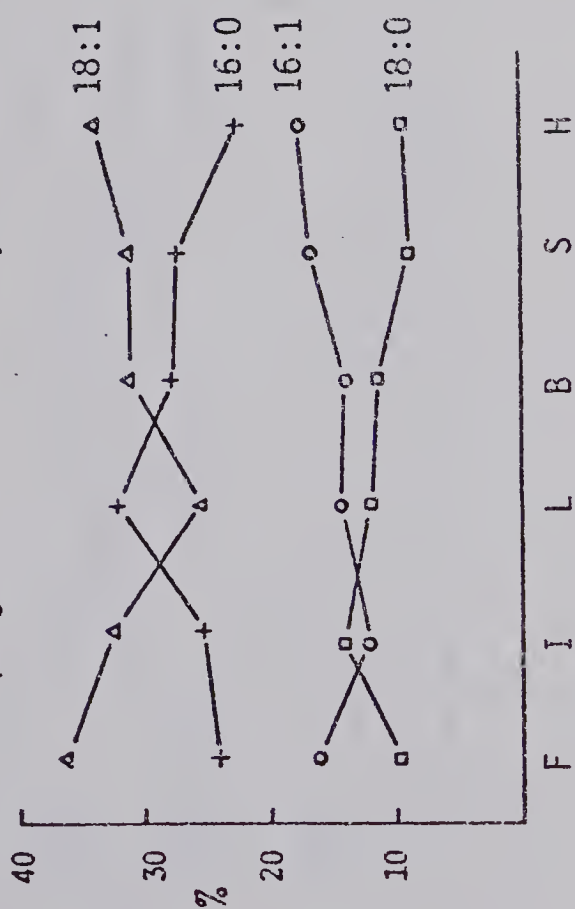


Figure 4. Variation in elk muscle triacylglycerol fatty acid composition with anatomical location. Letters along the abscissas represent the following: F = extensor carpi radialis, I = infraspinatus, L = longissimus dorsi, B = biceps femoris, S = semitendinosus, H = long digital extensor and peroneus tertius. Fatty acids are represented by the following: + = 16:0, O = 16:1, □ = 18:0, △ = 18:1.

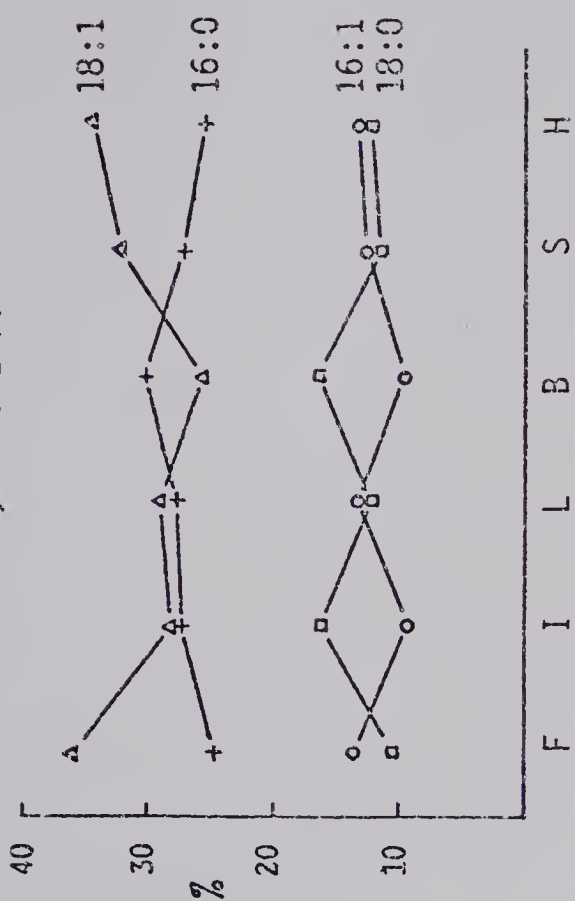
Animal No. ¹	Description
401	non-pregnant adult female, Mar. 1977
400	adult male, Mar. 1977
403	adult male, Jan. 1977
402	pregnant(?) adult female, Jan. 1977
404	pregnant(?) adult female, Dec. 1976
408	pregnant adult female, May 1977
407	calf male, Dec. 1976

¹ correspond to animal numbers in Appendix I.

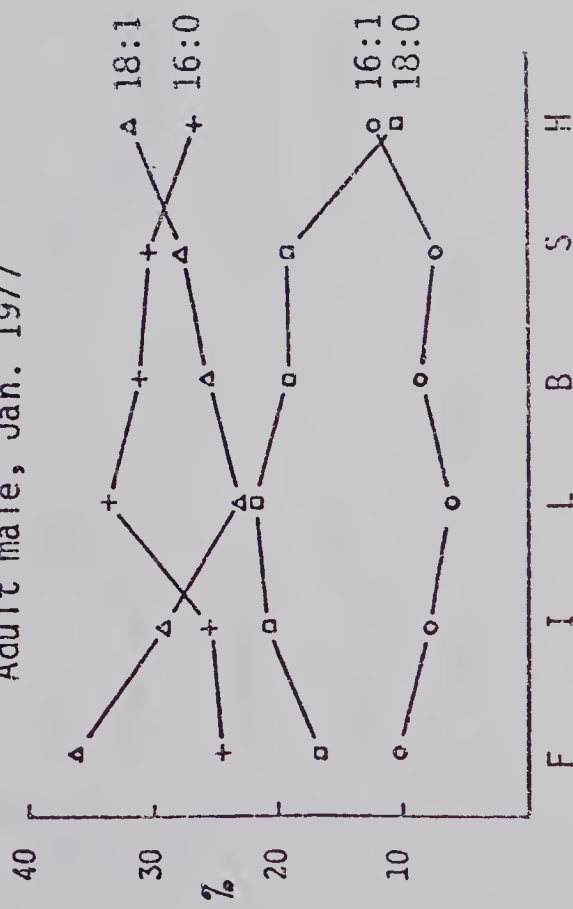
Non-pregnant adult female, Mar. 1977



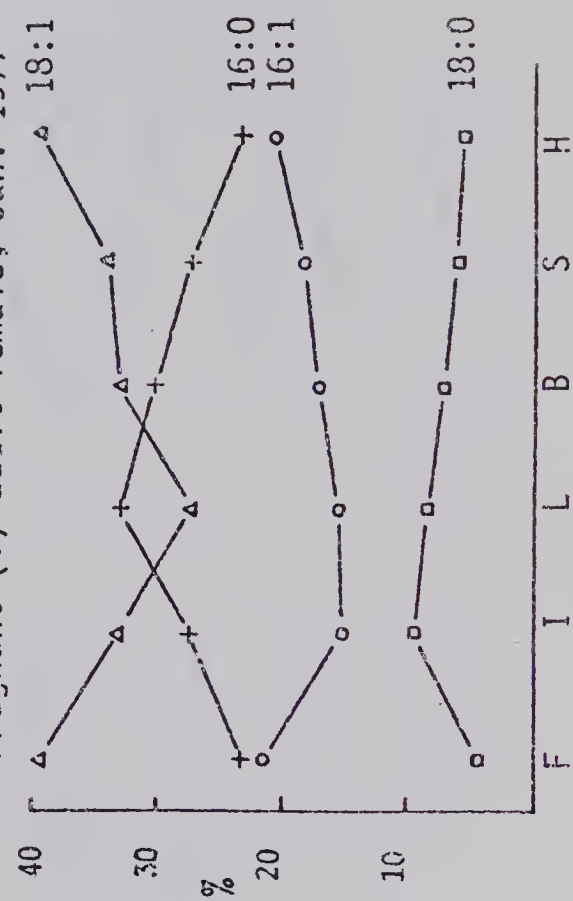
Adult male, Mar. 1977

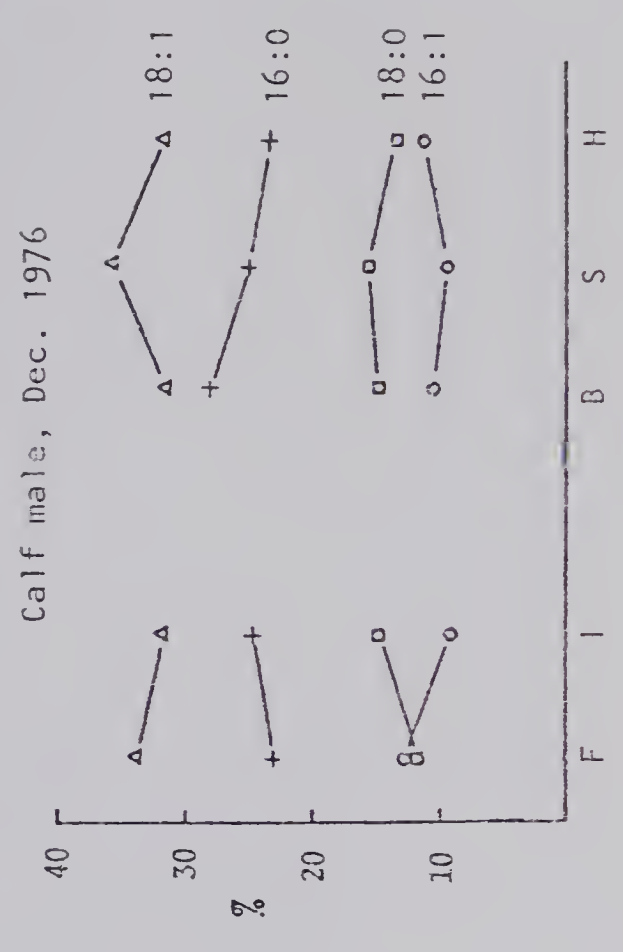
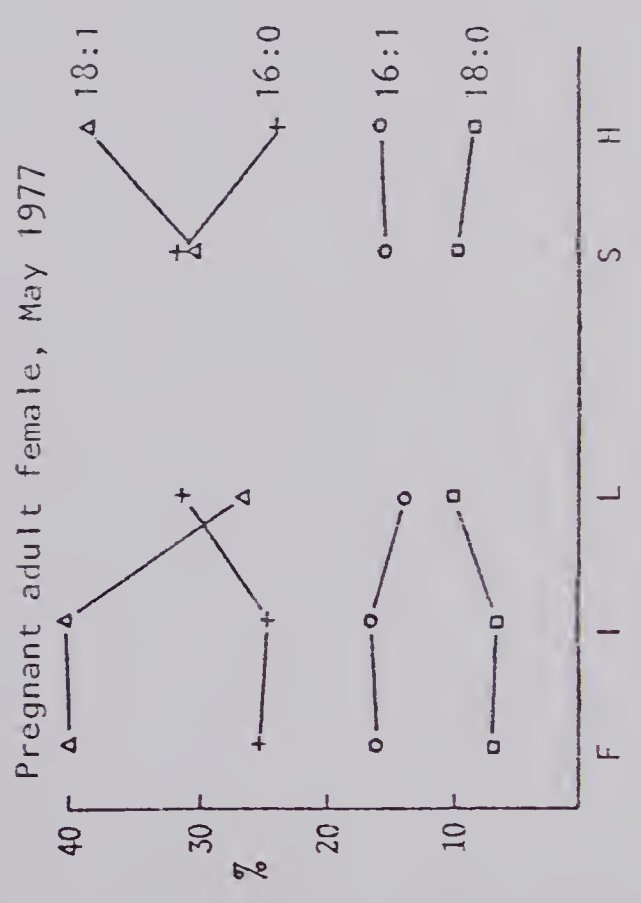
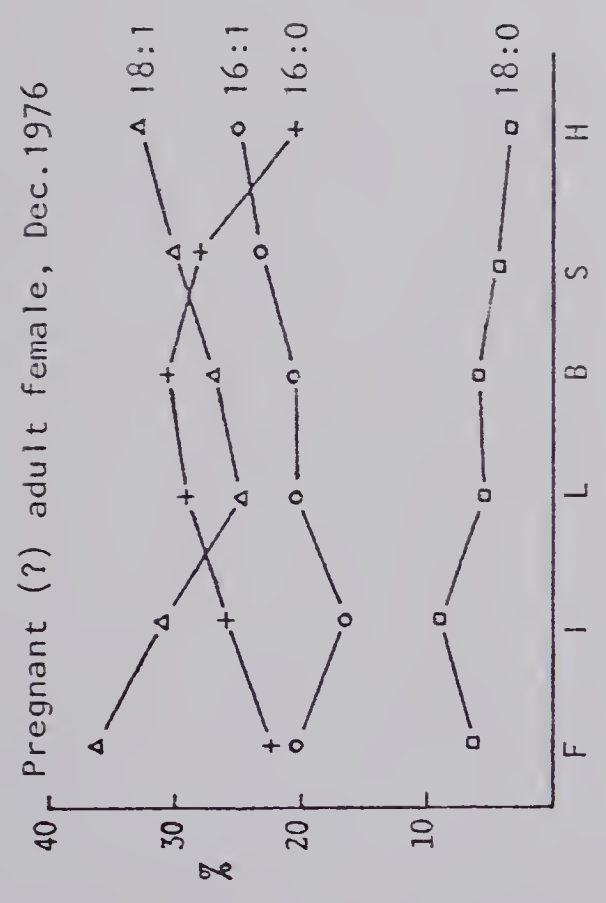


Adult male, Jan. 1977



Pregnant (?) adult female, Jan. 1977





1. The first part of the paper discusses the importance of the study of the history of the world, and the role of the world in the development of the human race. It is shown that the world is a complex system, and that the study of its history is essential for understanding the present and the future.

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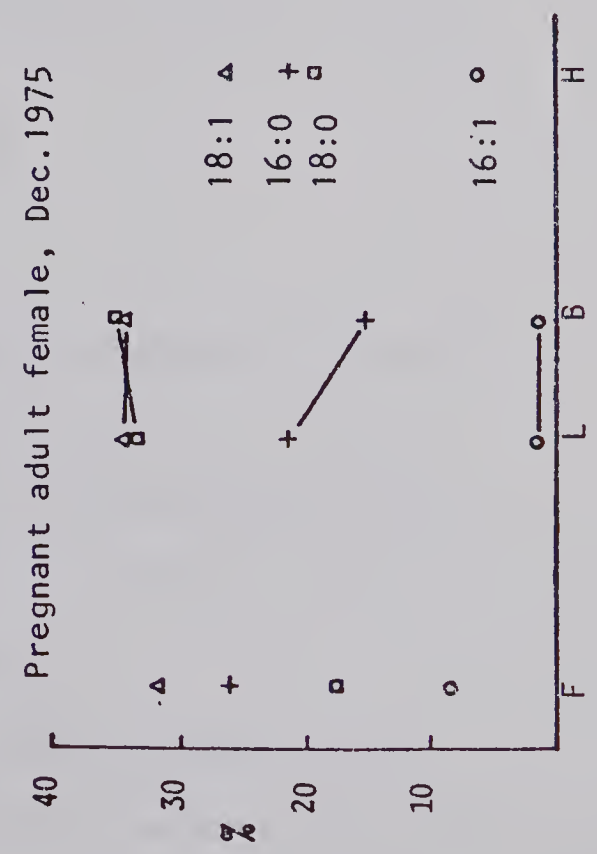
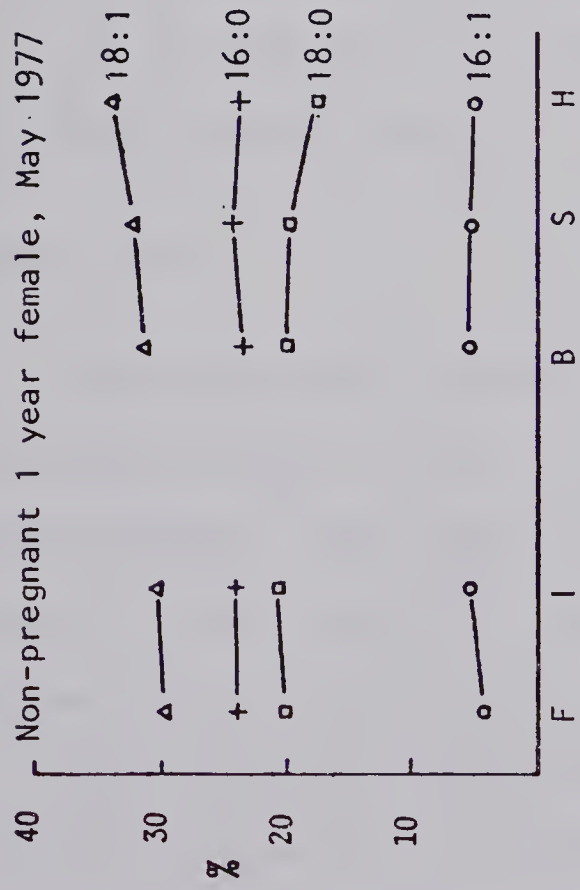
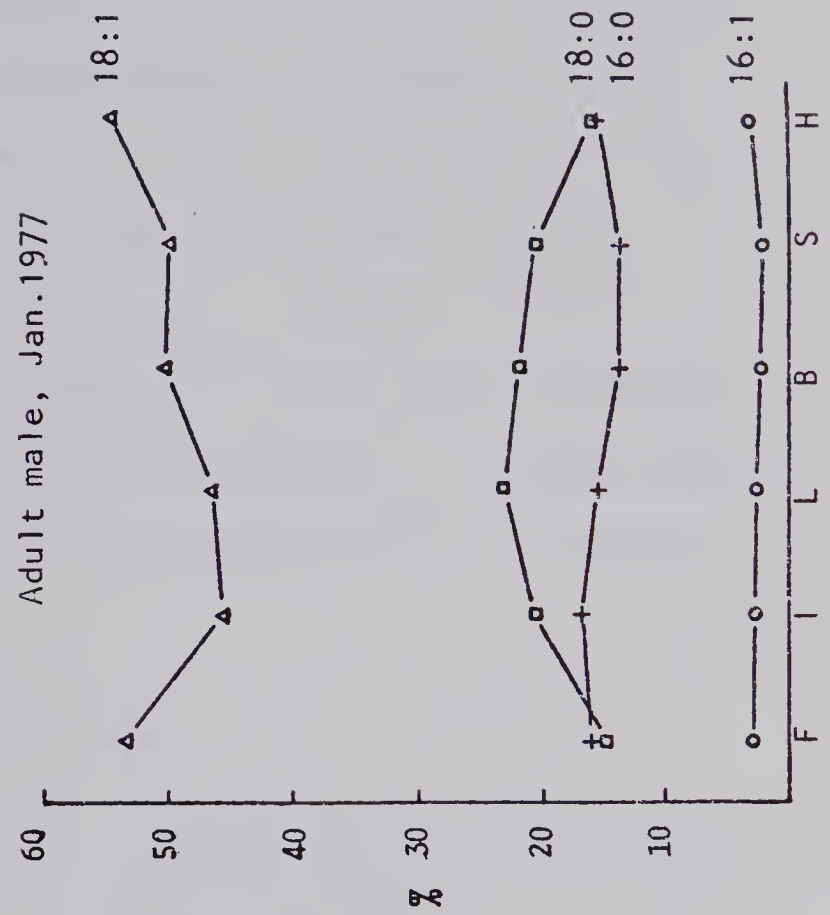
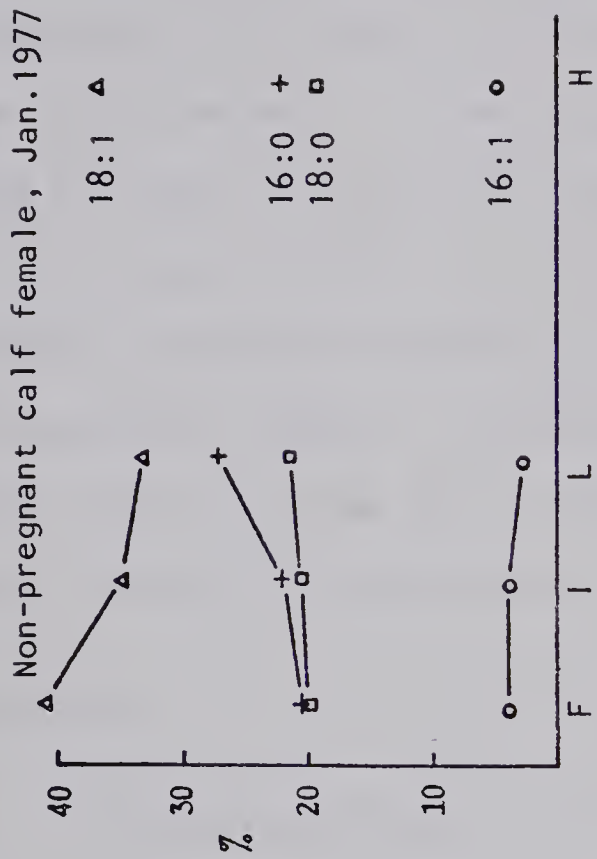
3. The third part of the paper discusses the importance of the study of the history of the world, and the role of the world in the development of the human race. It is shown that the world is a complex system, and that the study of its history is essential for understanding the present and the future.

4. The fourth part of the paper discusses the importance of the study of the history of the world, and the role of the world in the development of the human race. It is shown that the world is a complex system, and that the study of its history is essential for understanding the present and the future.

Figure 5. Variation in moose muscle triacylglycerol fatty acid composition with anatomical location. Letters along the abscissas represent the following: F = extensor carpi radialis, I = infraspinatus, L = longissimus dorsi, B = biceps femoris, S = semitendinosus, H = long digital extensor and peroneus tertius. Fatty acids are represented by the following: † = 16:0, ○ = 16:1, □ = 18:0, △ = 18:1.

Animal No. ¹	Description
409	non-pregnant 1 year female, May 1977
411	non-pregnant calf female, Jan. 1977
224	pregnant adult female, Dec. 1975
406	adult male, Jan. 1977

¹ corresponds to animal numbers in Appendix 1.



various anatomical locations were obtained from five white-tailed deer (Figure 6). The values for 16:0 and 16:1 show no noticeable trends in any of the animals. In the two deer collected during the winter there was a tendency for the extremities to have lower values for 18:0 and higher values for 18:1 than the other muscles. However, in the other animals, except the male fawn, the patterns for these two acids showed no particular trends. In the male fawn the 18:0 and 18:1 values showed only slight variation with anatomical location but only three muscles were available for comparison.

Antelope

The single antelope examined for the effect of anatomical location (Figure 7) showed little variation in values for either 16:0 or 16:1. Muscles from the extremities tended to have less 18:0 and more 18:1 than the other muscles did. The infraspinatus muscle had the largest 18:0 and lowest 18:1 values.

Bighorn sheep

Four muscles from a single bighorn sheep were compared (Figure 7). There was extremely little variation in either 16:0 or 16:1 values between muscles. The relative concentration of 18:1 was found to be greater in the l. dorsi while the concentration of 18:0 was found to be lower in the peripheral lower hind leg muscles.

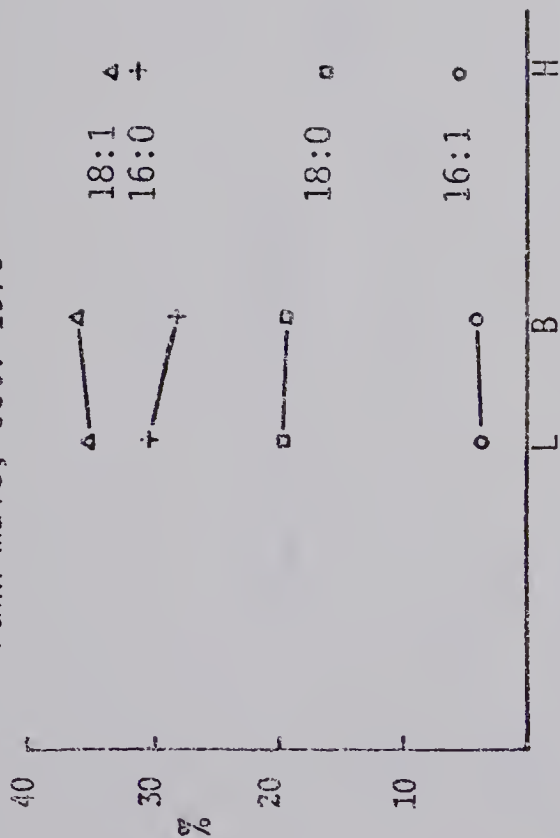
Many studies on domestic animals have shown that depot fat TG fatty acid composition varies with anatomical location with the inner depot fat containing a greater proportion of the saturated fatty acids than the more peripheral depots (Duncan and Garton, 1967; Chacko and Perkins, 1965; Pothoven et al., 1974). Garton and Duncan (1971) and Garton

Figure 6. Variation in white-tailed deer muscle triacylglycerol fatty acid composition with anatomical location. Letters along the abscissas represent the following: F = extensor carpi radialis, I = infraspinatus, L = longissimus dorsi, B = biceps femoris, S = semitendinosus, H = long digital extensor and peroneus tertius. Fatty acids are represented by the following: \dagger = 16:0, \circ = 16:1, \square = 18:0, \triangle = 18:1.

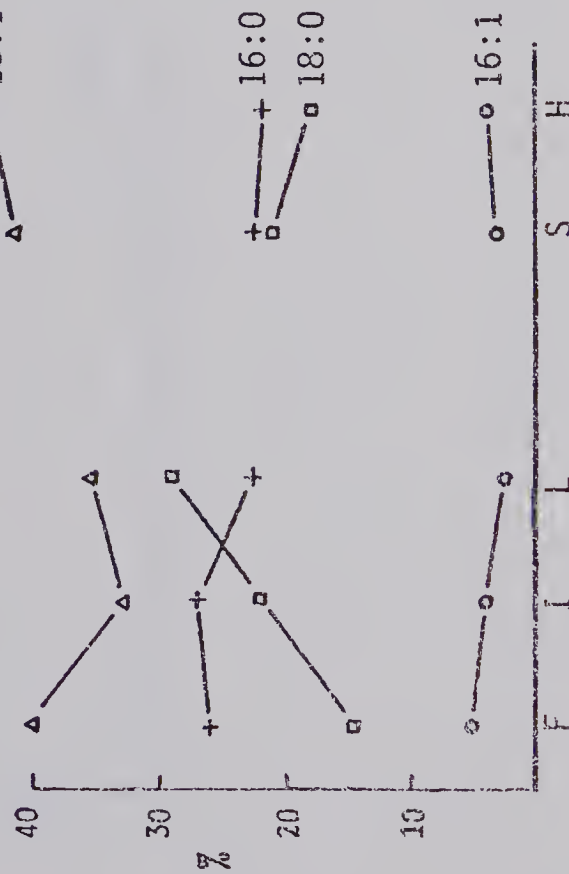
Animal No. ¹	Description
226	fawn male, Oct. 1975
416	non-pregnant fawn female, Feb. 1977
414	non-pregnant 1 year female, May 1977
415	adult male, Jan. 1977
412	adult male, July 1977

¹ correspond to animal numbers in Appendix 1.

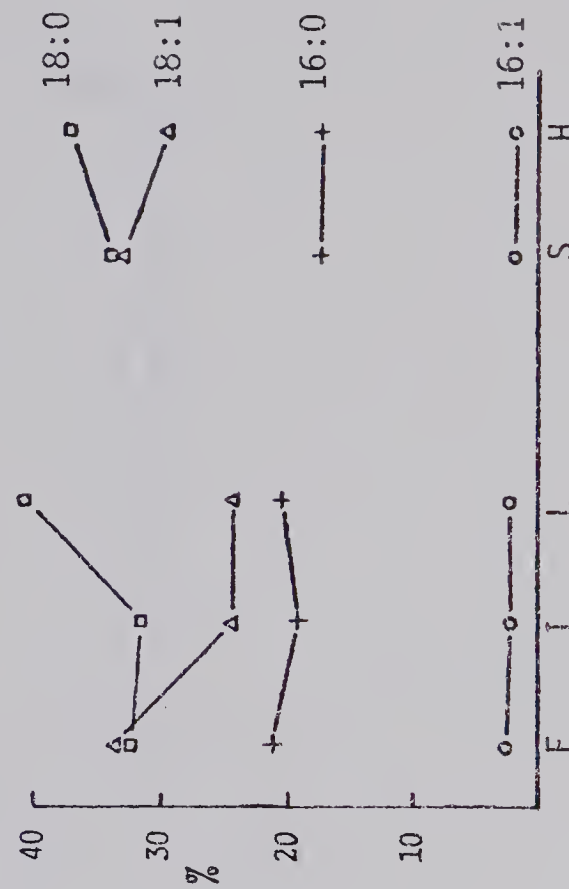
Fawn male, Oct. 1975



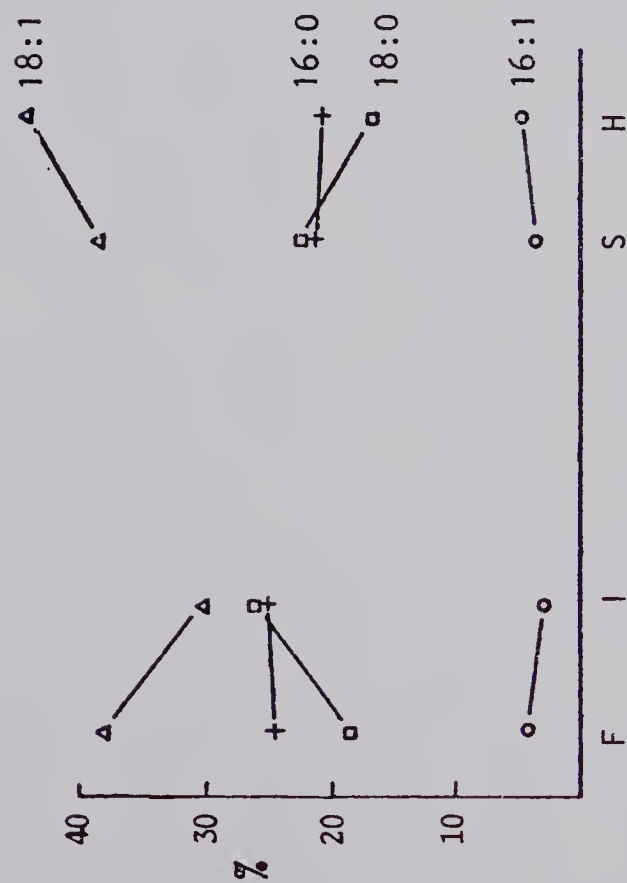
Non-pregnant fawn female, Feb. 1977



Non-pregnant, 1 year female, May 1977



Adult male, Jan. 1977



Adult male, July 1977

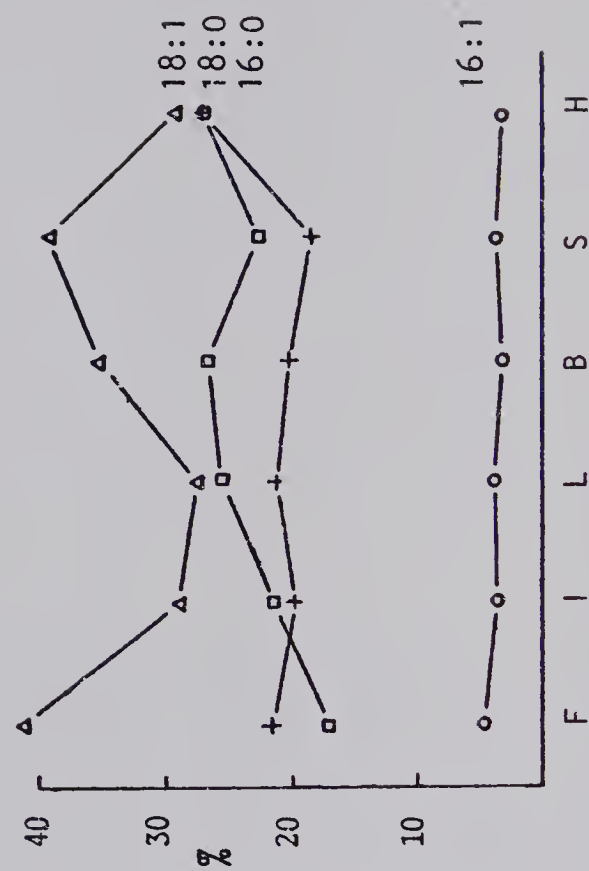
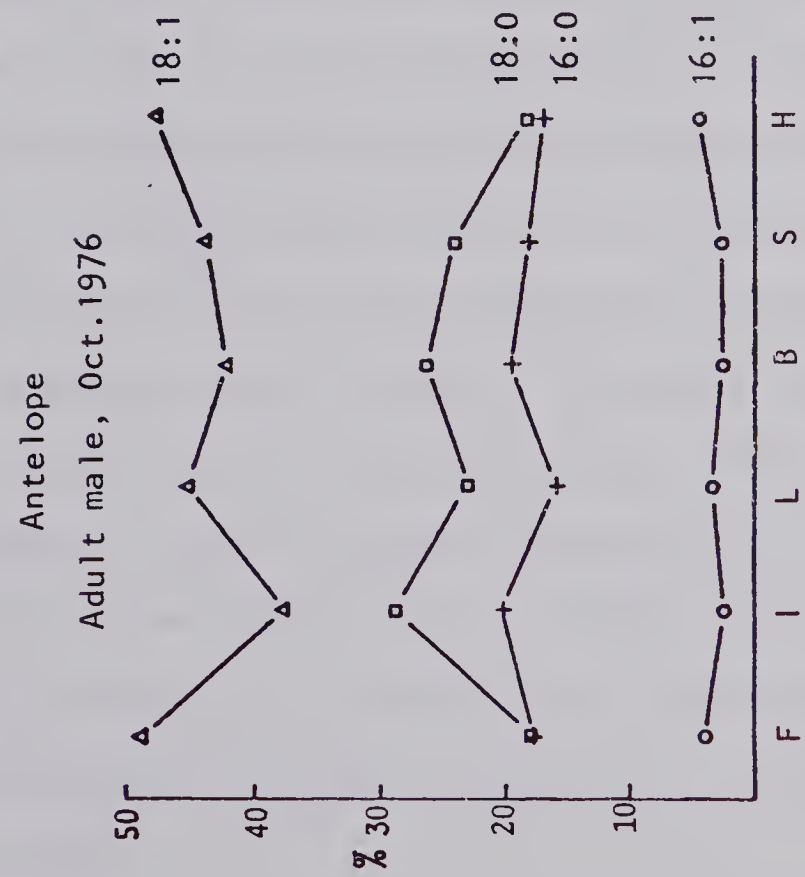
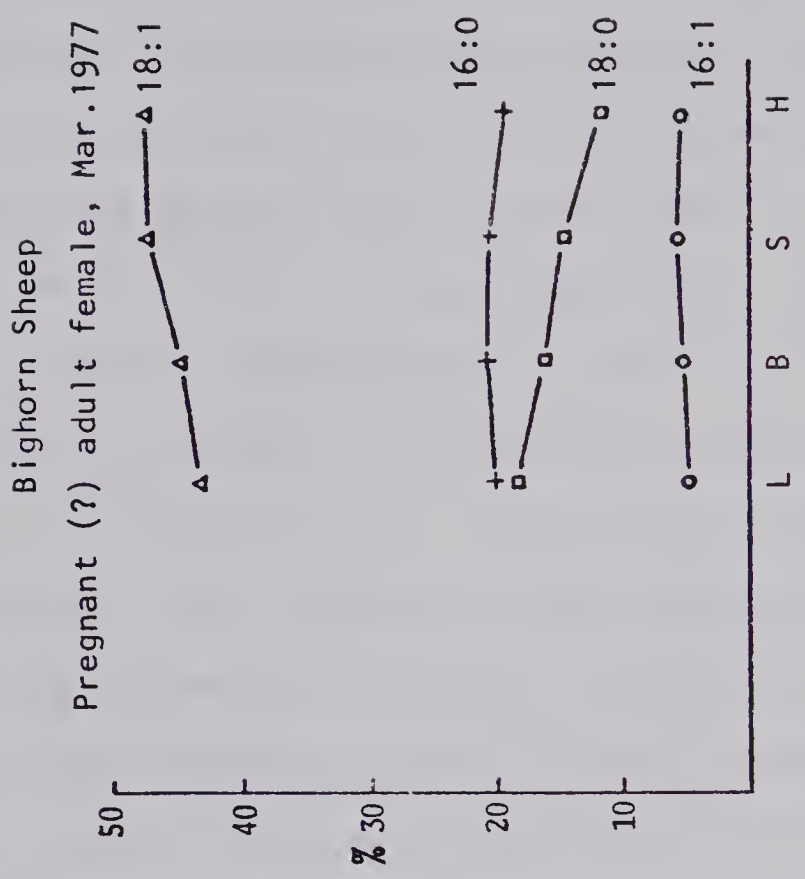


Figure 7. Variation in antelope and bighorn sheep muscle triacylglycerol fatty acid composition with anatomical location. Letters along the abscissas represent the following: F = extensor carpi radialis, I = infraspinatus, L = longissimus dorsi, B = biceps femoris, S = semitendinosus, H = long digital extensor and peroneus tertius. Fatty acids are represented by the following: + = 16:0, O = 16:1, □ = 18:0, △ = 18:1.

Animal No. ¹	Description
234	Antelope, adult male, Oct. 1976
235	Bighorn sheep, pregnant(?) adult female, Mar. 1977.

¹ correspond to animal numbers in Appendix 1.



et al. (1971) also found this for depot fats in wild ruminants. In a study of the fatty acid composition of various tissues from moose Tanhuanpää and Pulliainen (1975) found the same relationship for perinephric and subcutaneous adipose tissue for both total lipid and the TG fraction. For the PL fraction, however, they found a reverse pattern. For the two lipid fractions and for total lipid the degree of saturation in samples of hind leg muscle was less than in either the perinephric or subcutaneous tissues. Gillis et al. (1973) found bovine intramuscular (l. dorsi and biceps femoris) and subcutaneous lipid to have similar degrees of saturation. Marchello et al. (1970) compared the fatty acid composition of the l. dorsi, semimembranosus and triceps brachii muscles from steers and heifers for both chloroform-methanol and ether extracts. With the chloroform-methanol extract the l. dorsi of both steers and heifers had significantly more 18:0 than the other two muscles which had similar amounts. This also appeared to be true for the ether extract although this was not mentioned in their paper. In a similar study O'Keefe et al. (1968) examined the fatty acid composition of the TG and PL fractions of bovine l. dorsi, semitendinosus and triceps brachii muscles. The only highly significant difference recorded between muscles was that PL 18:0 was lower in the l. dorsi than in either of the other two muscles. O'Keefe et al. (1968) cite a study on beef by Hidaka et al. (1965) which reported there are no significant differences in the fatty acid composition of neutral fat or PL due to anatomical location of the muscle. The muscles they compared were not noted.

In the present study, none of the species examined showed consistent trends in PL fatty acid values. For TG however, some species

tended to demonstrate a decrease in the relative amount of saturated fatty acids when meat samples were collected from more peripheral parts of the animal. In elk there was a tendency for 16:0 to decrease and 18:1 to increase and in moose for 18:0 to decrease in the extremities. In the two white-tailed deer collected during winter there was a tendency for the extremities to have lower values for 18:0 and higher values for 18:1 than the more central muscles did. The single big-horn sheep also exhibited this trend for 18:0 and 18:1. The fatty acid values of the antelope and the remaining white-tailed deer did not exhibit any particular trends with anatomical location. The trends that were observed in this study are similar to those reported by others discussed above for adipose tissue in that when variations with anatomical location occurred they were consistent with increased unsaturation in peripheral tissues. Some studies on lipids that have demonstrated an increase in the degree of fatty acid unsaturation in peripheral tissues have suggested that this change is due to an adaptation to the low temperatures to which these tissues are exposed. A comparison of animals collected at different times of the year in the present study does not appear to support this. Most of the studies reporting changes in fatty acid composition with anatomical location compared internal depot fat with subcutaneous depot fat. One would not expect different muscles to show as much variation as seen with depot fat because muscles are more uniformly protected from cold. Still unexplained, however, are the apparently contradictory findings on variation between muscles and between muscle and depot fat found in the studies mentioned above.

In summary the relative amounts of individual PL fatty acids showed little variation with anatomical location but the degree of unsaturation in the TG fraction tended to be higher in peripheral muscles.

CHAPTER 4 Species classification of meat samples

INTRODUCTION

One of the major problems facing those involved in the enforcement of laws pertaining to the harvesting of wildlife is the positive identification of the species of origin of tissue samples. If the carcass, or parts thereof, cannot be identified with respect to species it may not be possible to convict an individual who has illegally harvested an animal.

Fatty acid values of muscles from a number of different anatomical locations were used to evaluate the forensic capabilities of fatty acid analysis for species determination. Muscles from a variety of anatomical locations were used because one cannot always identify the muscle(s) being submitted for analysis and anatomical locations can have some effect on fatty acid values.

EXPERIMENTAL

The methods of sample preparation and fatty acid analysis are outlined in Chapter 1. The classification analysis used is outlined in Klecka (1975). A classification function was derived for each species being compared. Each function is comprised of coefficients for each of the variables used in the discriminant analysis. The raw values for the variables are multiplied by the corresponding coefficient and the products are summed. Each sample is classified into the species with the highest probability of inclusion as determined by the classification function scores.

Following the preliminary analysis in which all six species are

compared one could use the combined results of the TG and PL analyses to eliminate some species from contention. For example, if an unknown is identified as being from an elk it can only be elk or moose. A further comparison using only these two species could then be done. The discrimination would theoretically be improved because the program does not have to compromise in its discrimination of these two species in order to optimize the discrimination of all species. Although the possibilities of misclassifying certain species may change with increased sample sizes the value of this approach to species identification will be explored using the information derived from this study.

RESULTS AND DISCUSSION

Details on the fatty acid composition of each muscle sample used in the analysis are tabulated in Appendix 1. The classification functions that were derived are presented in Appendix 2A. Results of the classification of meat sample TG by species are shown in Table 14. The overall success rate of correct classification was 77%. The prediction results show that for elk forty-eight (94%) samples were correctly classified while one sample (2%) was placed in each of the moose, white-tailed deer and antelope groups. For moose thirty-two samples (73%) were correctly classified while seven (16%) were called white-tailed deer, two (4%) were called antelope, two were called bighorn sheep and one (2%) was classified as an elk. Classification of white-tailed deer was not very successful with only thirty-three (58%) being correctly classified. Twelve (21%) of the whitetail samples were classified as mule deer, eight (14%) were called moose and four were called bighorn sheep. Contrary to the situation for white-tailed deer where a large

Table 14. Species classification of meat samples from all six species: predicted versus actual group membership.

<u>Triacylglycerols</u>							
Actual Species	No. of Cases	Predicted Species					
		Elk	Moose	W.T.D.	M.D.	Ant.	B.H.S.
Elk	51	48(94%)	1(2%)	1(2%)	0	1(2%)	0
Moose	44	1(2%)	32(73%)	7(16%)	0	2(4%)	2(4%)
W.T.D.	57	0	8(14%)	33(58%)	12(21%)	0	4(7%)
M.D.	17	0	1(6%)	1(6%)	14(82%)	0	1(6%)
Ant.	12	0	1(8%)	2(17%)	0	9(75%)	0
B.H.S.	12	0	0	0	0	0	12(100%)

Percent of cases correctly classified = 77%

<u>Phospholipids</u>							
Actual Species	No. of Cases	Predicted Species					
		Elk	Moose	W.T.D.	M.D.	Ant.	B.H.S.
Elk	50	43(86%)	5(10%)	1(2%)	0	1(2%)	0
Moose	37	3(8%)	33(89%)	0	0	1(3%)	0
W.T.D.	46	0	8(17%)	31(67%)	7(15%)	0	0
M.D.	16	0	0	3(19%)	11(69%)	0	2(12%)
Ant.	12	0	2(17%)	0	0	10(83%)	0
B.H.S.	12	0	0	0	0	0	12(100%)

Percent of cases correctly classified = 81%.

percentage were misclassified as the closely related mule deer, only one (6%) mule deer was called a whitetail. Of the remaining mule deer samples one was called a moose, one a bighorn sheep and fourteen (82%) were classified correctly. Nine (75%) of the antelope samples were correctly classified while one (8%) was called a moose and two (17%) were called white-tailed deer. All twelve of the bighorn sheep samples were classified correctly.

Results for the classification of samples by species using PL fatty acid values are also shown in Table 14. The overall rate of correct classification was 81%. The correct classification rate for elk and mule deer was lower using PL fatty acids than it was for TG fatty acids. The other species have a higher correct classification rate for PL than they did with TG. For elk forty-three (86%) of the samples were correctly classified while five (10%) were called moose, one (2%) was called a white-tailed deer and one an antelope. Thirty-three (89%) of the moose samples were classified correctly while three (8%) were called elk and one (3%) was called an antelope. Thirty-one (67%) of the white-tailed deer samples were correctly classified, seven (15%) were called mule deer and eight (17%) were called moose. The success of classifying mule deer samples was about the same as for white-tailed deer with eleven (69%) classified correctly, three (19%) classed as white-tailed deer and two (12%) as bighorn sheep. Ten (83%) of the twelve antelope samples were classified correctly while the other two (17%) were called moose. As with TG fatty acids all twelve bighorn sheep samples were correctly classified.

White-tailed deer was the only species for which the same muscle sample was misclassified using both TG and PL fatty acid values and this

occurred for six samples. Three of these were three different muscles from a single animal.

Elk

From the initial analyses using all six species it appears that samples classified as elk are most likely elk but they may also be misclassified moose. Further analyses comparing only these two species (Table 15) still does not give a clear discrimination of the two. For the TG fatty acid values in both the initial analysis with all six species and the subsequent analysis of only moose and elk the same sample from each species remained misclassified. For PL fatty acid values the comparison of elk and moose alone resulted in one elk and two moose samples being misclassified. This elk sample had also been misclassified in the initial analysis. In the analysis of elk and moose alone the samples that were misclassified using TG fatty acid values were not misclassified when PL fatty acids were used.

Moose

A meat sample classified as being from a moose in the comparison of all six species may also be a misclassified elk, white-tailed deer or antelope. An analysis of only these four species (Table 16) seems to provide no further improvement in correctly classifying samples classified as moose in the initial analysis. A comparison of moose and antelope only (Table 17) results in misclassification both ways (1 moose, 2 antelope) for TG fatty acids but with PL fatty acids none of the antelope samples were misclassified as moose. The one moose sample that was misclassified using PL fatty acids was not the same

Table 15. Species classification of meat samples from elk and moose: predicted versus actual group membership.

<u>Triacylglycerols</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Elk	Moose
Elk	51	50(98%)	1(2%)
Moose	44	1(2%)	43(98%)

Percent of cases correctly classified: 98%

<u>Phospholipids</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Elk	Moose
Elk	50	49(98%)	1(2%)
Moose	37	2(5%)	35(95%)

Percent of cases correctly classified: 96%

Table 16. Species classification of meat samples from elk, moose, white-tailed deer and antelope: predicted versus actual group membership.

<u>Triacylglycerols</u>					
Actual Species	No. of Cases	Predicted Species			
		Elk	Moose	W.T.D.	Ant.
Elk	51	49(96%)	1(2%)	1(2%)	0
Moose	44	1(2%)	34(77%)	8(18%)	1(2%)
W.T.D.	57	0	8(14%)	48(84%)	1(2%)
Ant.	12	0	2(17%)	2(17%)	8(67%)

Percent of cases correctly classified: 85%

<u>Phospholipids</u>					
Actual Species	No. of Cases	Predicted Species			
		Elk	Moose	W.T.D.	Ant.
Elk	50	42(84%)	5(10%)	1(2%)	2(4%)
Moose	37	2(5%)	32(86%)	2(5%)	1(3%)
W.T.D.	46	0	8(17%)	38(83%)	0
Ant.	12	0	2(17%)	0	10(83%)

Percent of cases correctly classified: 84%

Table 17. Species classification of meat samples from moose and antelope: predicted versus actual group membership.

<u>Triacylglycerols</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Moose	Ant.
Moose	44	43(98%)	1(2%)
Ant.	12	2(17%)	10(83%)

Percent of cases correctly classified: 95%

<u>Phospholipids</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Moose	Ant.
Moose	37	36(97%)	1(3%)
Ant.	12	0	12(100%)

Percent of cases correctly classified: 98%

one that was misclassified using TG fatty acids. As mentioned above a comparison of moose and elk alone resulted in misclassification of samples from both species for both TG and PL fatty acids. This was also the case when moose and white-tailed deer were compared (Table 18). In this comparison the only samples misclassified on the basis of both TG and PL fatty acid values were three muscles from an individual white-tailed deer. The net result of the above comparisons is that some samples of elk and white-tailed deer may be misclassified as moose.

White-tailed deer

A meat sample classified in the initial analysis as being from a white-tailed deer could be an elk, white-tailed deer or mule deer. Further comparison of only these three species (Table 19) seems to give no further insight into establishing the correct identity of the sample. Comparison of white-tailed deer with elk only (Table 20) resulted in all of the white-tailed deer samples being correctly classified for both TG and PL fatty acids. However, in both cases one elk sample was misclassified. These samples were from two different animals. Comparison of white-tailed deer and mule deer (Table 21) gave misclassifications of samples from both species for both the TG and PL analyses. Two of the white-tailed deer samples were misclassified for both TG and PL fatty acid values. These samples were from two different animals. From the results of the initial and subsequent species comparisons it was found that some elk and mule deer samples may be misclassified as white-tailed deer.

Table 18. Species classification of meat samples from moose and white-tailed deer: predicted versus actual group membership.

<u>Triacylglycerols</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Moose	W.T.D.
Moose	44	36 (82%)	8 (18%)
W.T.D.	57	9 (16%)	48 (84%)

Percent of cases correctly classified: 83%

<u>Phospholipids</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Moose	W.T.D.
Moose	37	36 (97%)	1 (3%)
W.T.D.	46	6 (13%)	40 (87%)

Percent of cases correctly classified: 92%

Table 19. Species classification of meat samples from elk, white-tailed deer and mule deer: predicted versus actual group membership.

<u>Triacylglycerols</u>				
Actual Species	No. of Cases	<u>Predicted Species</u>		
		Elk	W.T.D.	M.D.
Elk	51	50(98%)	1(2%)	0
W.T.D.	57	0	39(68%)	18(32%)
M.D.	17	0	3(18%)	14(82%)

Percent of cases correctly classified: 82%

<u>Phospholipids</u>				
Actual Species	No. of Cases	<u>Predicted Species</u>		
		Elk	W.T.D.	M.D.
Elk	50	49(98%)	1(2%)	0
W.T.D.	46	0	38(83%)	8(17%)
M.D.	16	0	4(25%)	12(75%)

Percent of cases correctly classified: 88%

Table 20. Species classification of meat samples from elk and white-tailed deer: predicted versus actual group membership.

<u>Triacylglycerols</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Elk	W.T.D.
Elk	51	50(98%)	1(2%)
W.T.D.	57	0	57(100%)

Percent of cases correctly classified: 99%

<u>Phospholipids</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		Elk	W.T.D.
Elk	50	49(98%)	1(2%)
W.T.D.	46	0	46(100%)

Percent of cases correctly classified: 99%

Table 21. Species classification of meat samples from white-tailed deer and mule deer: predicted versus actual group membership.

<u>Triacylglycerols</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		W.T.D.	M.D.
W.T.D.	57	42(74%)	15(26%)
M.D.	17	4(24%)	13(76%)

Percent of cases correctly classified: 74%

<u>Phospholipids</u>			
Actual Species	No. of Cases	<u>Predicted Species</u>	
		W.T.D.	M.D.
W.T.D.	46	39(85%)	7(15%)
M.D.	16	4(25%)	12(75%)

Percent of cases correctly classified: 82%

Mule deer

If a meat sample was classified as originating from a mule deer in the initial analysis of all six species the meat sample was from either a mule deer or a white-tailed deer. As mentioned above, further comparisons of these two species alone still results in misclassification of some samples from both species (Table 21).

Antelope

Samples classified as antelope in the comparison of all six species were from either elk, moose or antelope. Further analysis with only bighorn sheep left out (Table 22) resulted in moose samples being the only ones misclassified as antelope for both TG and PL fatty acids. As mentioned previously comparison of these two species alone still resulted in moose being misclassified as antelope (Table 17).

Bighorn sheep

In the analysis of the samples from all six species mule deer was the only species for which samples were misclassified as bighorn sheep for both TG and PL fatty acids. Further analysis with only these two species (Table 23) resulted in all of the samples being correctly classified for TG and PL fatty acid values. It was therefore possible to correctly classify all samples classified in the initial analysis as bighorn sheep.

From the classification results for discriminant analysis it was only possible to get a positive identification for bighorn sheep samples. Unfortunately, all of these samples were from adult females from a

Table 22. Species classification of meat samples from elk, moose, white-tailed deer, mule deer and antelope: predicted versus actual group membership.

<u>Triacylglycerols</u>						
Actual Species	No. of Cases	Predicted Species				
		Elk	Moose	W.T.D.	M.D.	Ant.
Elk	51	49(96%)	1(2%)	1(2%)	0	0
Moose	44	1(2%)	34(77%)	8(18%)	0	1(2%)
W.T.D.	57	0	8(14%)	31(54%)	16(28%)	2(4%)
M.D.	17	0	1(6%)	2(12%)	14(82%)	0
Ant.	12	0	1(8%)	3(25%)	0	8(67%)

Percent of cases correctly classified: 75%

<u>Phospholipids</u>						
Actual Species	No. of Cases	Predicted Species				
		Elk	Moose	W.T.D.	M.D.	Ant.
Elk	50	42(84%)	5(10%)	2(4%)	0	1(2%)
Moose	37	2(5%)	34(92%)	0	0	1(3%)
W.T.D.	46	0	8(17%)	31(67%)	7(15%)	0
M.D.	16	0	0	3(19%)	13(81%)	0
Ant.	12	0	2(17%)	0	0	10(83%)

Percent of cases correctly classified: 81%

Table 23. Species classification of meat samples from mule deer and bighorn sheep: predicted versus actual group membership.

Triacylglycerols

Actual Species	No. of Cases	Predicted Species	
		M.D.	B.H.S.
M.D.	17	17(100%)	0
B.H.S.	12	0	12(100%)

Percent of cases correctly classified: 100%

Phospholipids

Actual Species	No. of Cases	Predicted Species	
		M.D.	B.H.S.
M.D.	16	16(100%)	0
B.H.S.	12	0	12(100%)

Percent of cases correctly classified: 100%

single population, possibly limiting the strength of this observation.

A disadvantage of relying on the classification procedure used above is that one is not aware of the probability that a given meat sample has been correctly classified. In this classification procedure a meat sample is classified as originating from the particular species for which it has the highest probability of membership irregardless of the actual probability value. For example, the probabilities for which elk meat samples were correctly classified as being from elk varied from 1.000 to 0.510. It would be more informative in evaluating the classification results if the actual probabilities of group membership were known for each meat sample. The classification procedure used in this study provides this information and the probability of species membership of each of the meat samples used in this study is provided in Appendix 3A. If the highest probability is less than 1.000 the second highest is also provided.

Some apparent trends were noticed in the classification results. For TG fatty acids there seemed to be a sex effect in the classification of moose as the proportion of misclassified samples from females was three times that of samples from males. There appeared to be an effect of anatomical location in the classification of moose and white-tailed deer using TG fatty acids. For moose the proportion of l. dorsi samples misclassified was five times that of the other muscles combined. For white-tailed deer the situation was reversed and ten times as many samples from the 'other' muscles were misclassified than l. dorsi samples. Half of these misclassified 'other' muscles from white-tailed deer were samples of flank. The only trend noticed in misclassified PL samples was that five of the seven misclassified elk samples were

lower leg muscle.

These apparent trends suggest that if one has prior knowledge of factors such as sex or anatomical location the success of classification of unknowns would be improved by comparing them only with samples of similar background. An illustration of this is a comparison of the species classification results using all muscles (Table 14) versus the results using l. dorsi only (Table 24). With l. dorsi only there was an increase in the overall success rate for TG from 77% to 83% and for PL from 81% to 94% and there were fewer cases where species could not be distinguished.

Alternate analysis

Prior to doing the discriminant function analysis some intuitive methods were attempted based on certain trends that were observed in the raw data. For the TG fatty acids it was observed that elk tended to differ from the other species, most notably in the values for fatty acids 14:1, 16:1 and 18:1. One method used to exploit these trends was to express 18:1 as a percent of the sum of 14:1, 16:1 and 18:1. These results are shown in Figure 8. The most obvious feature of the figure is that while most elk samples have values between 50 and 80% samples from the other five species have, with two exceptions, values greater than 80%. Classification of samples using the midpoint between the mean for elk and the mean of each of the other species as the dividing line results in two elk samples being misclassified but no samples from the other species would be misclassified as elk. The upper 95% confidence limit for elk and the lower 95% confidence limits for the other species are indicated in Figure 8. One of the two elk

Table 24. Species classification of longissimus dorsi meat samples from all six species: predicted versus actual group membership.

Triacylglycerols

Actual Species	No. of Cases	Predicted Species					
		Elk	Moose	W.T.D.	M.D.	Ant.	B.H.S.
Elk	18	17(94%)	1(6%)	0	0	0	0
Moose	23	0	19(83%)	3(13%)	0	0	1(4%)
W.T.D.	20	0	1(5%)	15(75%)	3(15%)	1 (5%)	0
M.D.	8	0	0	1(12%)	6(75%)	0	1(12%)
Ant.	6	0	1(17%)	0	1(17%)	4(67%)	0
B.H.S.	7	0	0	0	0	0	7(100%)

Percent of cases correctly classified: 83%

Phospholipids

Actual Species	No. of Cases	Predicted Species					
		Elk	Moose	W.T.D.	M.D.	Ant.	B.H.S.
Elk	16	16(100%)	0	0	0	0	0
Moose	17	0	17(100%)	0	0	0	0
W.T.D.	13	0	2(15%)	9(69%)	2(15%)	0	0
M.D.	7	0	0	0	7(100%)	0	0
Ant.	6	0	0	0	0	6(100%)	0
B.H.S.	7	0	0	0	0	0	7(100%)

Percent of cases correctly classified: 94%

Figure 8. Species comparison using muscle triacylglycerol fatty acids:

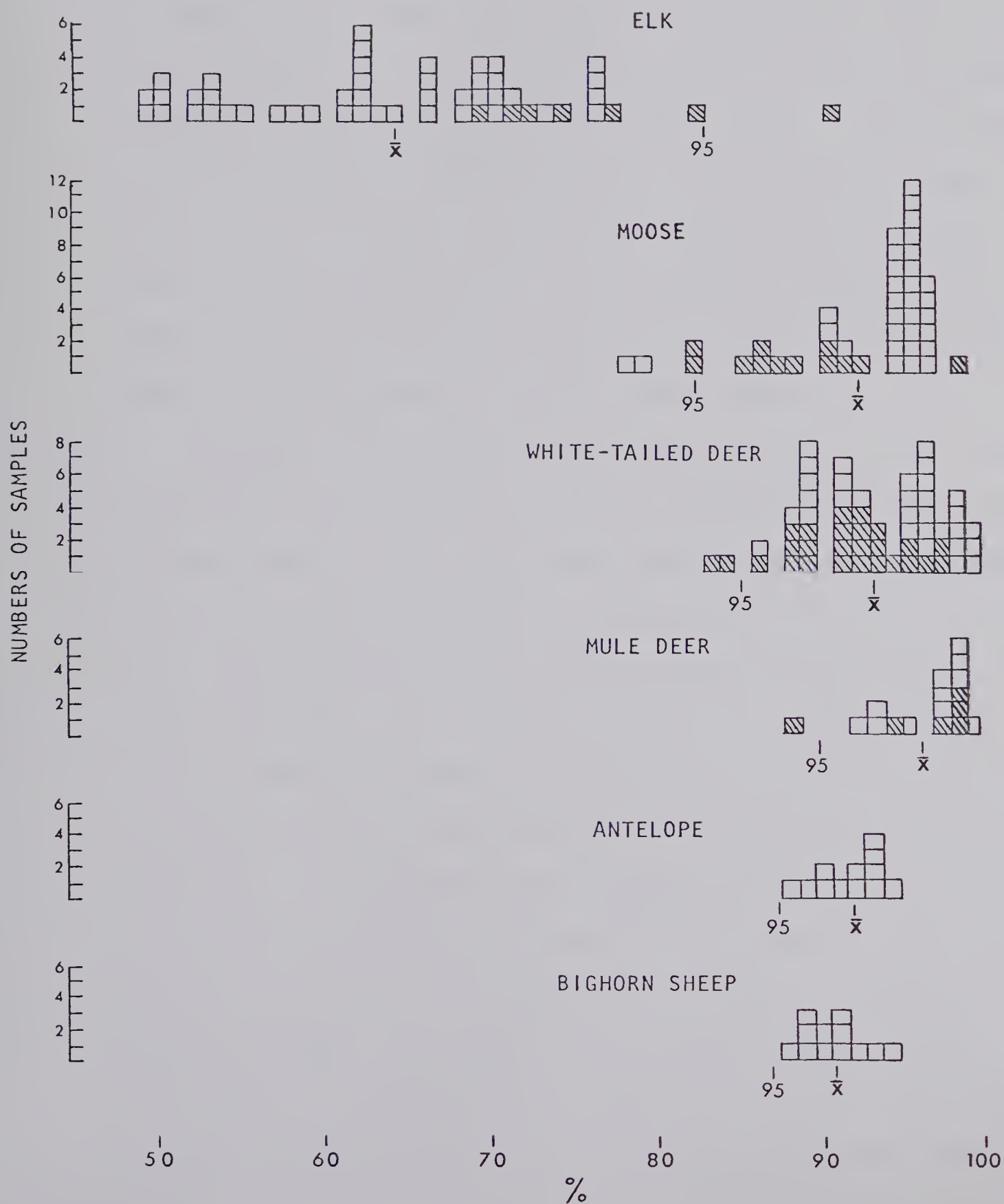
$$\frac{18:1}{14:1 + 16:1 + 18:1} \times 100\%$$

▨ = 4 to 15 month

□ = >15 months

\bar{x} = group mean

95 = 95% confidence limit



samples that would be misclassified using the medians of the elk mean and the mean of each of the other species as the dividing line falls outside the 95% confidence limit of elk and within the 95% limit of each of the other species. Four moose samples fall on or outside the 95% limit for that species and are within the 95% limit of elk. No samples of the other species fall within the elk 95% limit. The two elk samples that would be misclassified using the ratio method were also misclassified using discriminant analysis. These samples were from a male and a female both about eight months old. When only animals over 15 months of age are considered the 95% confidence limit for the ratio is 78.5% for elk and 84.6% for moose. So, if a sample is known to be from an adult there would be much less difficulty in distinguishing elk from the other species.

In summary, the overall success rates for species classification were 77% for the TG and 81% for the PL. Following the initial comparison each species in turn was further compared with only those species that had samples misclassified into that species in the initial comparison. This was done to try to improve the discriminations and this was usually achieved. Only for bighorn sheep was it possible to get positive identification of the samples but the strength of this is limited by the fact that all of the sheep samples were from adult females from a single population. It is suggested that the classification probabilities should be examined for evaluating the classification results. Some apparent trends in the classification results were noted which suggested that comparing unknowns with knowns from the same anatomical location or from animals of the same sex for example would increase the probability for correct classification. It was noted that elk tended to differ

from the other species in the values for TG 14:1, 16:1 and 18:1. A comparison of the species using the ratio $(18:1/14:1 + 16:1 + 18:1) \times 100\%$ was used to illustrate this.

CHAPTER 5 Age and sex classification of meat samples from elk, moose and white-tailed deer.

INTRODUCTION

From a forensic standpoint it would be valuable to be able to discriminate between meat samples from animals less than six months old, seven to fifteen months old, and greater than fifteen months old, since there are different regulations governing the hunting of animals in these age groups. There are also different restrictions on the harvesting of male and female animals that are at least one year of age. Therefore it is often necessary to determine the age and/or sex of a carcass or part thereof. The present chapter evaluates the reliability of using muscle fatty acid composition for such determinations.

EXPERIMENTAL

The methods of sample preparation and fatty acid analysis are outlined in Chapter 1. Functions for the classification of elk, moose and white-tailed deer by age and by sex were derived from the same manner as outlined in Chapter 4 for the classification of species. A variety of different muscles were used for the analyses for the same reason outlined in the previous chapter. Insufficient samples of mule deer, antelope and bighorn sheep were available for age or sex analysis. For elk, moose and white-tailed deer intraspecific comparisons were made between animals that were seven to fifteen months old and animals older than fifteen months. For the analysis of white-tailed deer, samples from several animals four to six months old were also used.

Classification by sex was limited to samples from animals that were known to be at least one year old.

RESULTS AND DISCUSSION

Details on the fatty acid composition of each muscle sample used in the analysis is tabulated in Appendix 1.

Age

The classification functions that were derived are presented in Appendix 2B. The results of the age classification of the elk meat samples are shown in Table 25. All eleven samples from seven to fifteen month old elk were correctly classified on the basis of their TG fatty acid pattern while six of the forty-four samples from animals in the older age group were misclassified. For PL two of the seven samples from animals in the younger age group and six of the forty-three from the older age animals were misclassified. Overall, 88% of the TG samples and 84% of the PL samples were correctly classified. The samples which were misclassified using the TG fraction were not the same ones that were misclassified using the PL fraction.

Table 26 shows the results for the classification of moose meat samples according to animal age. Classification of samples from animals in the seven to fifteen month group was very successful with all eleven samples being correctly classified for both TG and PL fatty acids. However, two of the thirty-three TG samples and one of the twenty-six PL samples from the older animals were misclassified. The overall rate for correct classification was 95% for TG and 97% for PL. The single meat sample that was misclassified using PL fatty acid values was correctly classified when TG fatty acid values were used.

The results for the classification of white-tailed deer meat samples by age are shown in Table 27. When TG fatty acid values were

Table 25. Classification by age of meat samples from elk: predicted versus actual group membership.

<u>Triacylglycerols</u>			
Actual Age	No. of Cases	Predicted Age	
		7 to 15 months	>15 months
7 to 15 months	7	7(100%)	0
>15 months	44	6(14%)	38(86%)

Percent of cases correctly classified: 88%

<u>Phospholipids</u>			
Actual Age	No. of Cases	Predicted Age	
		7 to 15 months	>15 months
7 to 15 months	7	5(71%)	2(29%)
>15 months	43	6(14%)	37(86%)

Percent of cases correctly classified: 84%

Table 26. Classification by age of meat samples from moose: predicted versus actual group membership.

<u>Triacylglycerols</u>			
Actual Age	No. of Cases	Predicted Age	
		7 to 15 months	>15 months
7 to 15 months	11	11(100%)	0
>15 months	33	2(6%)	31(94%)
Percent of cases correctly classified: 95%			

<u>Phospholipids</u>			
Actual Age	No. of Cases	Predicted Age	
		7 to 15 months	>15 months
7 to 15 months	11	11(100%)	0
>15 months	26	1(4%)	25(96%)
Percent of cases correctly classified: 97%			

Table 27. Classification by age of meat samples from white-tailed deer: predicted versus actual group membership.

<u>Triacylglycerols</u>				
Actual Age	No. of Cases	Predicted Age		
		4 to 6 months	7 to 15 months	>15 months
4 to 6 months	11	10(91%)	1(9%)	0
7 to 15 months	16	0	12(75%)	4(25%)
>15 months	30	1(3%)	4(13%)	25(83%)

Percent of cases correctly classified: 82%

<u>Phospholipids</u>				
Actual Age	No. of Cases	Predicted Age		
		4 to 6 months	7 to 15 months	>15 months
4 to 6 months	5	5(100%)	0	0
7 to 15 months	13	0	13(100%)	0
>15 months	28	0	6(21%)	22(79%)

Percent of cases correctly classified: 87%

used one of the eleven samples from the four to six month old animals was misclassified as belonging to the seven to fifteen month group. The remaining samples from the four to six month group were classified correctly. Four of the sixteen samples from the seven to fifteen month group and four of the thirty samples from the greater than fifteen month group were classified as belonging to the opposite group. In addition, one of the samples from the oldest age group was classified as belonging to the four to six month group. When PL fatty acid values were used all five samples from animals four to six months old and all thirteen samples from the seven to fifteen month group were correctly classified. Six of the twenty-eight samples from animals in the oldest age group were classified as belonging to the seven to fifteen month group. The overall correct classification rate was 82% for TG fatty acid values and 87% when PL fatty acid values were used. None of the samples that were misclassified using TG fatty acids were misclassified when PL fatty acids were used.

A few apparent trends were seen in the misclassification of samples. For TG from elk and white-tailed deer the majority of the misclassified samples were from males. This suggests that sex may have an additional effect on the differences seen. For elk and moose, TG samples from animals over fifteen months old were misclassified as belonging to the seven to fifteen month group but the reverse did not occur. This was also true for moose and white-tailed deer PL samples. Most of these samples were from yearlings while ages for the others were uncertain. It may be that fatty acid composition is still changing when animals are yearlings and dividing samples at the fifteen month point is masking an age effect. Overall, leg muscle samples were most often misclassified

using TG fatty acids and l. dorsi samples were most often misclassified when PL fatty acids were used. Knowledge of factors such as sex of the animal and the actual muscle being analyzed would result in improved accuracy of classification.

Two elk each had two muscles misclassified using TG and this was also true for another elk for PL. The other misclassified elk samples each came from a different animal. Each of the misclassified moose samples came from different animals. For white-tailed deer there were two animals that each had three muscles misclassified using TG and one of these animals also had three muscles misclassified using PL. The remaining misclassified white-tailed deer samples each came from a different animal.

The only case for which the classification results were completely correct was for the four to six month white-tailed deer PL group. There were other cases where there was one way misclassification, for example, for white-tailed deer PL some samples from the greater than fifteen month group were put in the seven to fifteen month group but not vice-versa. In some situations this information could be just as useful as if there were no misclassifications. As mentioned in the discussion on species classification the probability of group membership value may be a more meaningful criterion in evaluating the identity of a sample rather than merely accepting the assigned group membership. The probabilities for age classification of each sample are shown in Appendix 3B.

Sex

The classification functions that were derived are presented in Appendix 2C. The results for the sex classification of elk samples

are shown in Table 28. For TG one of the fifteen samples from males was misclassified as were three of the twenty-nine samples from females. When PL were used all fifteen samples from males were correctly classified but six of the twenty-eight samples from females were misclassified. The percent of cases correctly classified was 91% when TG fatty acids were used and 86% for PL fatty acids. Two of the samples were misclassified for both TG and PL fatty acids.

Sex classification results for the moose samples are shown in Table 29. When TG fatty acids were used five of the sixteen samples from males and one of the seventeen samples from females were misclassified resulting in an overall correct classification rate of 82%. For the PL fatty acids all twelve samples from males and all fourteen samples from females were correctly classified.

In the classification of white-tailed deer by sex (Table 30) four of the twenty-one TG samples from males were misclassified but all ten samples from females were correctly classified. When PL fatty acid values were used three of the nineteen samples from males and one of the nine samples from females were misclassified. The rate of correct classification was 87% for TG and 86% for PL. Samples that were misclassified when TG fatty acids were used were correctly classified when PL fatty acids were used.

For elk TG one animal had two muscle samples misclassified. These same two muscles plus a third one from this animal were misclassified using PL fatty acid values. Another animal had two muscles misclassified using PL but the other misclassified elk samples each came from a different animal. Each of the misclassified moose TG samples came from different animals and this was also true for the white-tailed deer TG

Table 28. Classification by sex of meat samples from elk: predicted versus actual group membership¹.

<u>Triacylglycerols</u>			
<u>Actual Sex</u>	<u>No. of Cases</u>	<u>Predicted Sex</u>	
		<u>Male</u>	<u>Female</u>
Male	15	14(93%)	1(7%)
Female	29	3(10%)	26(90%)
Percent of cases correctly classified: 91%			

<u>Phospholipids</u>			
<u>Actual Sex</u>	<u>No. of Cases</u>	<u>Predicted Sex</u>	
		<u>Male</u>	<u>Female</u>
Male	15	15(100%)	0
Female	28	6(21%)	22(79%)
Percent of cases correctly classified: 86%			

¹ All samples are from animals over one year of age.

Table 29. Classification by sex of meat samples from moose: predicted versus actual group membership¹.

<u>Triacylglycerols</u>			
Actual Sex	No. of Cases	<u>Predicted Sex</u>	
		Male	Female
Male	16	11(69%)	5(31%)
Female	17	1(6%)	16(94%)
Percent of cases correctly classified: 82%			

<u>Phospholipids</u>			
Actual Sex	No. of Cases	<u>Predicted Sex</u>	
		Male	Female
Male	12	12(100%)	0
Female	14	0	14(100%)
Percent of cases correctly classified: 100%			

¹ All samples are from animals over one year of age.

Table 30. Classification by sex of meat samples from white-tailed deer: predicted versus actual group membership¹.

Triacylglycerols

Actual Sex	No. of Cases	Predicted Sex	
		Male	Female
Male	21	17(81%)	4(19%)
Female	10	0	10(100%)

Percent of cases correctly classified: 87%

Phospholipids

Actual Sex	No. of Cases	Predicted Sex	
		Male	Female
Male	19	16(84%)	3(16%)
Female	9	1(11%)	8(89%)

Percent of cases correctly classified: 86%

¹ All samples are from animals over one year of age.

and PL samples.

Only for moose was it possible to correctly classify all samples. For elk one could be sure of the classification for samples classified as female and for white-tailed deer for samples classified as male. Most of the misclassified elk samples were from females while the majority of misclassified moose and white-tailed deer samples were from males. The only other trend noticed for misclassification was that when TG fatty acids were used all of the misclassified moose samples were from l. dorsi muscle.

As for species and age classifications the probability of group membership value may be an important criterion in evaluating the sex classification of a sample. The probabilities of group membership for samples classified by sex are presented in Appendix 3C.

In summary, the success rates for classification by age ranged from 82% for white-tailed deer TG to 97% for moose PL. The only group for which the classification results were completely correct was for the four to six month white-tailed deer PL group. The success rates for classification by sex ranged from 82% for moose TG to 100% for moose PL. Only for moose PL was it possible to correctly classify all samples.

CHAPTER 6. The effect of cooking on the fatty acid composition of meat

INTRODUCTION

It is occasionally necessary to identify samples of cooked meat. Most techniques for analyzing uncooked samples would need to be modified to be useable on cooked samples. The majority of the reports on the effect of cooking on fatty acid composition of meat from domestic animals have reported that there are no significant effects (Chang and Watts, 1952; Giam and Dugan, 1965; Castledine and Davies, 1968; de Lumen et al., 1974). Likewise, Hubbard and Pocklington (1968) found commercial processing to have little effect on the fatty acid pattern of meat. Some authors, however have noted some limited effects of cooking on fatty acid composition. The present tests were done to see if cooking would result in an alteration of fatty acid pattern to the extent that it would affect the forensic usefulness of the technique.

EXPERIMENTAL

The effect of cooking on the fatty acid composition of muscle was determined by cooking muscle samples to an internal temperature of either 61 C or 77 C in an oven set at 193 C. The internal temperatures of the meat samples were monitored by thermocouples inserted into the middle of the samples. In trial 1, samples of the semitendinosus and l. dorsi muscles from one side of a mule deer carcass were used as the controls and samples from the corresponding muscles from the opposite side were cut into two equal parts and used for cooking. In trial 2 samples of the semitendinosus muscle from both sides of an elk carcass were cut into two equal parts with one part being used as the control

and the other part cooked. Following cooking, the samples were stored, prepared and analyzed in the same manner as outlined previously for uncooked samples.

RESULTS AND DISCUSSION

The results of the two cooking trials are shown in Tables 31 and 32. In each of the comparisons of corresponding cooked and uncooked meat samples most of the fatty acids showed some differences in their values. No consistent trends in these differences for any of the fatty acids were observed. The differences that were observed may be only a reflection of experimental error(s) and/or small differences in the initial fatty acid composition of the meat samples. The meat samples from the elk were all correctly classified as to species of origin for both TG and PL fatty acids. For the TG fatty acids all of the mule deer l. dorsi samples and the semitendinosus sample cooked to 61 C were classified as moose while the other two semitendinosus samples were classified as bighorn sheep. For PL fatty acids all of the mule deer samples were classified as antelope. Cooking, therefore, had no effect on the species classification of the elk meat samples and an inconsistent effect on the classification of the mule deer meat samples. As mentioned previously most studies have found cooking to have no significant effect on fatty acid composition but other studies have noted some limited effects. Campbell and Turkki (1967) found that PL 18:2 was higher in cooked pork than in raw pork. Terrel et al. (1968) found lower 18:3 in the neutral fraction of broiled bovine l. dorsi steaks than in uncooked steaks. The small differences between cooked and uncooked samples in the above studies would likely have little

Table 31. The triacylglycerol and phospholipid fatty acid composition of mule deer semitendinosus and longissimus dorsi muscles before and after cooking.

		<u>Triacylglycerols</u>					
		<u>Semitendinosus</u>			<u>Longissimus dorsi</u>		
		<u>Uncooked</u>	<u>61 C¹</u>	<u>77 C</u>	<u>Uncooked</u>	<u>61 C</u>	<u>77 C</u>
Fatty Acid	14:0	2.4	2.5	2.8	2.8	2.6	2.2
	14:1	0.4	0.5	0.5	0.6	0.8	0.3
	16:0	24.1	24.9	25.8	24.5	21.0	24.0
	16:1	5.1	5.1	5.0	4.6	3.6	3.9
	18:0	15.8	16.3	16.0	19.8	18.6	19.3
	18:1	41.5	41.9	41.3	38.2	37.0	38.3
	18:2	5.1	4.5	4.2	4.6	5.4	6.1
	18:3	0.4	0.3	0.3	0.3	1.1	0.4
	20:3	0.0	0.0	0.0	0.0	0.0	0.1
	20:4	0.5	0.2	0.2	0.2	0.3	1.4
	20:5	0.0	0.0	0.0	0.0	0.0	0.0
	22:5	0.0	0.0	0.0	0.0	0.0	0.0

		<u>Phospholipids</u>					
		<u>Semitendinosus</u>			<u>Longissimus dorsi</u>		
		<u>Uncooked</u>	<u>61 C</u>	<u>77 C</u>	<u>Uncooked</u>	<u>61 C</u>	<u>77 C</u>
Fatty Acid	14:0	0.3	0.3	0.3	0.9	1.0	0.1
	14:1	0.1	0.1	0.1	0.2	0.0	0.0
	16:0	15.4	18.5	15.1	16.1	20.5	17.4
	16:1	2.5	3.6	1.8	2.7	4.3	3.4
	18:0	14.5	14.5	16.4	14.2	13.5	13.9
	18:1	21.2	15.8	17.9	21.0	17.8	15.4
	18:2	20.9	23.0	26.0	20.9	20.2	25.7
	18:3	0.4	0.4	0.3	0.4	0.8	0.4
	20:3	0.9	1.2	0.9	1.1	0.5	1.1
	20:4	16.8	16.0	15.2	16.0	12.2	16.0
	20:5	0.7	0.8	0.6	0.7	0.6	0.8
	22:5	2.4	1.7	1.4	1.5	1.6	1.6

¹ internal temperature to which meat sample was heated.

Table 32. The triacylglycerol and phospholipid fatty acid composition of elk semitendinosus muscle before and after cooking.

		<u>Triacylglycerols</u>			
		<u>Uncooked</u>	<u>61 C¹</u>	<u>Uncooked</u>	<u>77 C</u>
Fatty Acid	14:0	4.3	4.0	4.8	4.7
	14:1	2.1	1.8	2.4	2.6
	16:0	31.2	31.0	31.4	31.2
	16:1	14.3	13.6	16.0	16.8
	18:0	10.3	10.5	8.9	8.4
	18:1	29.8	30.0	29.4	28.9
	18:2	3.6	3.8	3.1	3.6
	18:3	0.6	0.7	0.6	0.6
	20:3	0.1	0.1	0.1	0.1
	20:4	0.4	0.2	0.3	0.6
	20:5	0.0	0.0	0.0	0.0
	22:5	0.0	0.0	0.0	0.0
		<u>Phospholipids</u>			
		<u>Uncooked</u>	<u>61 C</u>	<u>Uncooked</u>	<u>77 C</u>
Fatty Acid	14:0	0.3	0.4	0.3	0.3
	14:1	0.4	0.5	0.4	0.4
	16:0	16.5	15.5	16.6	16.1
	16:1	3.7	3.6	3.5	3.8
	18:0	14.9	15.5	14.8	15.1
	18:1	12.9	13.1	12.3	13.1
	18:2	25.4	26.2	26.0	26.7
	18:3	3.3	3.2	3.0	3.3
	20:3	1.8	1.7	1.7	1.4
	20:4	11.5	10.7	11.7	11.5
	20:5	2.2	1.9	2.3	2.3
	22:5	3.3	3.1	3.1	2.7

¹ Internal temperature to which meat sample was heated.

effect on classification when the degree of variation in fatty acid composition of meat samples from different animals, different muscles, etc. is taken into consideration.

The method of cooking, temperature of cooking, and length of time the meat is cooked are all factors that could have some effect on changes in fatty acid composition due to cooking. The effects of these variables are unknown and extensive studies would need to be done to evaluate them before one could use fatty acid analysis of cooked meat for forensic purposes. The cooking conditions used in this experiment did not appear to affect fatty acid composition.

GENERAL SUMMARY AND CONCLUSIONS

A brief review of the various analytical techniques applicable to the characterization of wildlife tissues is provided prior to the presentation of material from the present study.

The purpose of the present study was to determine the fatty acid composition of skeletal muscle tissue from several wild ungulate species native to Alberta and to determine the applicability of these results to wildlife forensic studies. Upon initial inspection of the data it was apparent that elk had a relatively unique pattern of fatty acids in their skeletal musculature. The triacylglycerols (TG) of the longissimus dorsi (l. dorsi) muscle of elk contained higher concentrations of 14:0, 14:1, 16:0 and 16:1 and lower concentrations of 18:0 and 18:1 than did the other animals. Discriminant analysis of l. dorsi samples by species revealed that all species pairs had significantly different ($P < .01$) TG and phospholipid (PL) fatty acid compositions except for the white-tailed deer - mule deer TG pair ($P < .05$). Elk had the most distinctive fatty acid pattern while, as might be expected from phylogenetic relationships, white-tailed deer and mule deer were the most similar.

A possible explanation for the relatively unique TG fatty acid pattern of elk is that at some point in their evolutionary history the ancestors of elk experienced a mutation(s) resulting in a reduction in their ability to lengthen fatty acids 14 to 16 carbon atoms long. The fatty acid composition of abomasal contents from several elk, moose and white-tailed deer collected at various times of year were found to be similar in this study.

Therefore diet did not seem to be a major factor in determining the species differences in muscle fatty acid composition.

Comparison of l. dorsi samples by animal age was done for elk, moose and white-tailed deer. There were no consistent differences between age groups among the three species but there were some trends within and between species for some of the individual fatty acids. Elk and moose fetal (approximately half term) samples had similar values for most of the TG and PL fatty acids. In the present study the fetal muscle concentrations of the PL polyunsaturated fatty acids 20:4 and 22:5 were similar to those in adult skeletal muscle while the concentration of their 18-carbon precursors, particularly 18:2, were extremely low. During the post-natal growth of elk the relative concentrations of TG 14:1 and 16:1 increased and 18:1 decreased. For elk PL there was a tendency for 16:0, 16:1 and 18:1 to increase and for 18:2 and 20:5 to decrease. Moose TG 14:0 decreased and 18:1 increased during post natal growth but PL fatty acids showed little change. There were minimal differences in TG and in PL fatty acids of white-tailed deer with age.

The effect of sex on the fatty acid composition of l. dorsi from elk, moose and white-tailed deer was examined. For each of the species the two sexes had overlapping ranges in values for most of the fatty acids and no clear differences due to sex were observed.

Variation in fatty acid pattern with anatomical location of muscle was examined in a number of individual elk, moose, white-tailed deer, antelope and bighorn sheep. Phospholipid fatty acids showed little

variation with anatomical location. Variations in some TG fatty acids occurred among muscles for each animal examined but the variations were not consistent for all individuals. There was a trend for TG fatty acids of the more peripheral muscles to be less saturated. This trend agrees with previous reports on depot fats. One would not expect to see as great a difference between different muscles as has been reported for internal and subcutaneous depot fats because muscles have a relatively more uniform thermal environment.

Fatty acid values for muscles from a variety of different anatomical locations were used to derive classification functions for the classification of samples by species and for the classification of elk, moose and white-tailed deer by age and by sex. For species classification the overall rates of correct classification using TG and PL were 77% and 81% respectively. After the initial classification of samples each species was compared with only those species that had samples misclassified into that species. Elk and moose could not be completely distinguished and in addition moose could not be completely distinguished from white-tailed deer. Some elk and mule deer samples were misclassified as white-tailed deer while the only species that could not be distinguished from mule deer was white-tailed deer. Moose was the only species from which antelope could not be completely distinguished. It was possible to correctly classify all the bighorn sheep samples.

Examination of the raw data revealed that elk tended to have more 14:1 and 16:1 and less 18:1 than the other species. It was felt that expressing 18:1 as a percentage of the sum of the three fatty acids might provide a useful method for identifying elk. Except for two elk calves all of the elk samples had values between 50% and 80% while all

samples of the other species except two moose had values greater than 80%. Classification of samples using the midpoint between the means of each species as the dividing line would result in the two elk calves being misclassified but no samples from the other species would be misclassified as elk.

For classification by age, the samples were placed in one of three age groups: 1) four to six months, 2) seven to fifteen months, and 3) greater than 15 months. Samples from animals in the youngest age group were only available for white-tailed deer. The success rates for classifying samples by age ranged from 82% for white-tailed deer TG to 97% for moose PL. For elk TG all samples in the seven to fifteen month group were correctly classified and this was also true for moose for TG and PL and for white-tailed deer PL. In addition, for white-tailed deer TG no four to six month samples were classified as being greater than fifteen months and no seven to fifteen month samples were classified as being four to six months and for PL all four to six month samples were classified correctly and none of the greater than fifteen month group were classified as being four to six months. For all other age group comparisons for the three species there were some misclassifications. For elk and moose TG and moose and white-tailed deer PL samples from animals over fifteen months old were misclassified as belonging to the seven to fifteen month group but the reverse did not occur. The fact that most of these samples were known yearlings suggested that fatty acid composition may still be changing when animals are yearlings and dividing samples at the fifteen month point masked an age effect.

Classification by sex was done for elk, moose and white-tailed

deer over one year of age. The overall success rate varied from 82% for moose TG to 100% for moose PL. All male elk samples and all moose samples were classified correctly using the PL fatty acid pattern and all female white-tailed deer were correctly classified using the TG fatty acid pattern. There were misclassifications for all other comparisons.

A disadvantage in relying on the classification results is that a sample would be classified the same regardless of whether the probability of it belonging to a particular group was 51% or 100%. It would therefore be more informative to also consider the actual probability of group membership in deciding the identity of a sample.

Certain types of samples seemed to be more prone to misclassification in some of the comparisons. Because of this it would tend to become easier to evaluate the reliability of a classification the more the amount of background data that is available on a sample.

The effect of cooking on muscle fatty acid composition was investigated. Cooking meat samples to internal temperatures of 61 C or 77 C in an oven at 193 C did not affect TG or PL fatty acid composition.

The results of this study indicate that fatty acid analysis of meat may be a useful forensic tool in wildlife enforcement particularly for the identification of elk meat. Although other techniques such as electrophoresis may be more accurate for species determination, fatty acid analysis of meat may be a useful adjunct for determination of species as well as age and sex. It must be kept in mind that this investigation was an exploratory one and that for most of the analyses there were some limitations in the sampling.

REFERENCES CITED

- Aberle, E. D., and R. A. Merkel. 1966. Solubility and electrophoretic behavior of some proteins of post-mortem aged bovine muscle. *J. Fd. Sci.* 31: 151-156.
- Adjutantis, G., and A. Coutselinis. 1972. Estimation of the time of death by potassium levels in the vitreous humour. *Forens. Sci.* 1: 55-60.
- Awad, A., W. D. Powrie, and O. Fennema. 1968. Chemical deterioration of frozen bovine muscle at -4°C . *J. Fd. Sci.* 33: 227-235.
- Beattie, K. H., R. H. Giles Jr., and C. J. Cowles. 1977. Lack of research in wildlife law enforcement. *Wildl. Soc. Bull.* 5: 170-174.
- Bhatia, R. Y. P. 1974. Specificity of some plant lectins in the differentiation of animal blood. *Forens. Sci.* 4: 47-52.
- Bird, G. W. G. 1954. Observations on the interactions of the erythrocytes of various species with certain seed agglutinins. *Br. J. Exp. Path.* 35: 252-254.
- Blonde, D. J., E. J. Kresack, and G. W. Kosicki. 1967. The effects of ions and freeze-thawing on supernatant and mitochondrial malate dehydrogenase. *Can. J. Biochem.* 45: 641-650.
- Booren, A., R. A. Field, and J. E. Kunsman Jr. 1973. Carbonyl and fatty acid analysis of antelope and beef fat. *J. Fd. Sci.* 38: 63-65.
- Bourque, B. J., K. Morris, and A. Spiess. 1978. Determining the season of death of mammal teeth from archeological sites: a new sectioning technique. *Science* 199: 530-531.
- Brüggeman, J., U. Drescher-Kaden, R. Schubert, H. Erbersdobler, and D. Giesecke. 1975. Comparative study in reindeer and white-tailed deer of Finland on rumen metabolism and fatty acids of adipose tissues. In: *Proceedings of The First International Reindeer/Caribou Symposium*, pp. 290-296.
- Brunetti, O. A., K. F. Levine, and J. D. Banks. 1977. Laboratory aids to wildlife law enforcement - wildlife physical evidence. In: *Forensic Science: Symposium Proceedings*. Alberta Recreation, Parks and Wildlife, pp. 21-38.
- Brush, A. H. 1976. Waterfowl feather proteins: analysis of use in taxonomic studies. *J. Zool., Lond.* 179: 467-498.
- Buchanan, B. C. 1971. New techniques in meat identification. In: *Transactions 28th Northeast Fish and Wildlife Conference*, pp. 27-31.

- Bunch, T. D., R. W. Meadows, W. C. Foote, L. N. Egbert, and J. J. Spillett. 1976. Identification of ungulate hemoglobins for law enforcement. *J. Wildl. Manage.* 40: 517-522.
- Burnham, J. T., J. Preston-Burnham, and C. R. Fontan. 1976. The state of the art of bone identification by chemical and microscopic methods. *J. Forens. Sci.* 21: 340-342.
- Butcher, P. D., and C. M. Hawkey. 1977. Haemoglobins and erythrocyte sickling in the Artiodactyla: a survey. *Comp. Biochem. Physiol.* 57A: 391-398.
- Calaprice, J. R. 1970. A preliminary report on x-ray spectrometric analysis and discrimination of salmonids from different geographic areas. Fisheries Research Board of Canada. Technical Report Number 200, 35 pp.
- _____. 1971. X-ray spectrometric and multivariate analysis of sockeye salmon (*Oncorhynchus nerka*) from different geographic regions. *J. Fish. Res. Bd. Canada* 28: 369-377.
- Campbell, A. M., and P. R. Turkki. 1967. Lipids of raw and cooked ground beef and pork. *J. Fd. Sci.* 32: 143-146.
- Castledine, S. A., and D. R. A. Davies. 1968. Aids to the identification of meat in meat products. *Assoc. Publ. Analy. J.* 6: 39-52.
- Chacko, G. K., and E. G. Perkins. 1965. Anatomical variation in fatty acid composition and triglyceride distribution in animal depot fats. *J. Amer. Oil Chem. Soc.* 42: 1121-1124.
- Chang, I. C. L., and B. M. Watts. 1952. The fatty acid content of meat and poultry before and after cooking. *J. Amer. Oil Chem. Soc.* 29: 334-338.
- Clemens, E., V. Arthaud, R. Mandigo, and W. Woods. 1973. Fatty acid composition of bulls and steers as influenced by age and dietary energy level. *J. Anim. Sci.* 37: 1326-1331.
- Connell, J. J. 1962. Changes in amount of myosin extractable from cod flesh during storage at -14° . *J. Sci. Fd. Agric.* 13: 607-617.
- Cowan, I. McT., and P. A. Johnston. 1962. Blood serum protein variations at the species and subspecies level in deer of the genus *Odocoileus*. *Systematic Zool.* 11: 131-138.
- Croonquist, D. A. 1977. New methods of species identification. 33rd Annual Conference. Association of Midwestern Fish and Game Law Enforcement Officers, pp. 100-103.
- Cummings, E. W. 1970. Techniques of game law enforcement using starch gel electrophoresis. M.Sc. Thesis. Univ. of Calif. (Davis). 53 pp.

- Darskus, R. L., and J. M. Gillespie. 1971. Breed and species differences in the hair proteins of four genera of Caprini. *Aust. J. Biol. Sci.* 24: 515-524.
- Davidson, W. M. 1966. Sexual dimorphism in nuclei of polymorphonuclear leukocytes in various animals. pp. 59-75. In: *The Sex Chromatin*. K. L. Moore ed. W. B. Saunders Co., Philadelphia, Pa.
- deLumen, B. O., V. C. Witte, and M. E. Bailey. 1974. Effects of processing on the major fatty acids of separable porcine tissues. I. Influence of roasting fresh pork. *J. Anim. Sci.* 39: 309-316.
- Denney, R. N. 1965. Sex determination in dressed elk carcasses. 23rd. N. Amer. Wildl. Conf., pp. 501-513.
- Dhindsa, D. S., T. H. Cochran, A. Castro, J. R. Swanson, and J. Metcalfe. 1975. Serum biochemical and electrophoretic values from four deer species and from pronghorn antelope. *Amer. J. Vet. Res.* 36: 1455-1457.
- Dilworth, T. G., and J. A. McKenzie. 1970. Attempts to identify meat of game animals by starch-gel electrophoresis. *J. Wildl. Manage.* 34: 917-921.
- Duncan, W. R. H., and G. A. Garton. 1967. The fatty acid composition and intramolecular structure of triglycerides derived from different sites in the body of the sheep. *J. Sci. Fd. Agric.* 18: 99-102.
- _____, E. R. Ørskov, and G. A. Garton. 1974. Effect of different dietary cereals on the occurrence of branched-chain fatty acids in lamb fats. *Proc. Nutr. Soc.* 33: 81A.
- Escoubas, J. R., J. J. Guenther, and K. K. Novotny. 1975. The effects of freezing and thawing on lactate dehydrogenase isoenzymes from bovine muscle. pp. 207-210 In: *Animal Sciences and Industry Research Report MP-94*. Oklahoma Agric. Exper. Station.
- Flynn, A., and W. Franzmann. 1977. Forensic mineral element analysis in moose management. In: 13th N. Amer. Moose Conference and Workshop, pp. 99-105.
- Folch, J., M. Lees, and G. H. S. Stanley. 1957. A simple method for the isolation and purification of total lipides from animal tissues. *J. Biol. Chem.* 226: 497-509.
- Franzmann, A. W., A. Flynn, and P. D. Arneson. 1975. Levels of some mineral elements in Alaskan moose hair. *J. Wildl. Manage.* 39: 374-378.
- _____, _____, _____. 1977. Alaskan moose hair element values and variability. *Comp. Biochem. Physiol.* 57(3A): 299-306.

- Fugate, H. G. Jr., and S. R. Penn. 1971. Immunodiffusion technique for the identification of animal species. *J. Assoc. Off. Anal. Chem.*: 1152-1156.
- Garton, G. A., and W. R. H. Duncan. 1971. Fatty acid composition and intramolecular structure of triglycerides from adipose tissue of the red deer and the reindeer. *J. Sci. Fd. Agric.* 22: 29-33.
- _____, _____, and E. H. McEwan. 1971. Composition of adipose tissue triglycerides of the elk (*Cervus canadensis*), caribou (*Rangifer tarandus groenlandicus*), moose (*Alces alces*), and white-tailed deer (*Odocoileus virginianus*). *Can. J. Zool.* 49: 1159-1162.
- Giam, I., and L. R. Dugan Jr. 1965. The fatty acid composition of free and bound lipids in freeze-dried meats. *J. Fd. Sci.* 30: 262-265.
- Gilbert, B. M. 1973. Definitive characteristics separating ungulate genera. pp. 37-39. In: B. M. Gilbert, *Mammalian Osteo-Archaeology: North America*. Special publications. Missouri Archeol. Soc., Columbia, Mo.
- Gill, J. D., and D. C. O'Meara. 1965. Estimating time of death in white-tailed deer. *J. Wildl. Manage.* 29: 471-486.
- Gillis, A. T., N. A. M. Eskin, and R. L. Cliplef. 1973. Fatty acid composition of bovine intramuscular and subcutaneous fat as related to breed and sex. *J. Fd. Sci.* 38: 408-411.
- Grunbaum, B. W. 1977. Electrophoresis in forensic applications. *Industrial Research*, 15 Nov.: 19, 13-19.
- Gustavsson, I., and C. O. Sundt. 1968. Karyotypes in five species of deer (*Alces alces* L., *Capreolus capreolus* L., *Cervus elaphus* L., *Cervus nippon nippon* Temm. and *Dama dama* L.). *Hereditas* 60: 233-248.
- Harris, M. J., T. H. J. Huisman, and F. A. Hayes. 1973. Geographic distribution of hemoglobin variants in the white-tailed deer. *J. Mammal.* 54: 270-274.
- Hay, J. D., R. W. Currie, and F. H. Wolfe. 1973. Polyacrylamide disc gel electrophoresis of fresh and aged chicken muscle proteins in sodium dodecylsulfate. *J. Fd. Sci.* 38: 987-990.
- Hecker, A. L., D. A. Cramer, and D. F. Hougham. 1975. Compositional and metabolic growth effects in the bovine. Muscle, subcutaneous and serum total fatty acids. *J. Fd. Sci.* 40: 144-149.
- Herin, R. A. 1968. Physiological studies in the rocky mountain elk. *J. Mammal.* 49: 762-764.

- Hidiroglou, M., and D. T. Spurr. 1975. Influence of cold exposure and diet change on the trace element composition of hair from shorthorn cattle. *Can. J. Anim. Sci.* 55: 31-38.
- Hildebrand, M. 1955. Skeletal differences between deer, sheep and goats. *Calif. Fish and Game* 41: 327-346.
- Hoekstra, T. W., and P. G. Carr. 1977. Sex determination in white-tailed deer tissues. In: *Forensic Science: symposium proceedings*. Alberta Recreation, Parks and Wildlife, pp. 212-232.
- Hood, R. L., and E. Allen. 1971. Influence of sex and postmortem aging on intramuscular and subcutaneous bovine lipids. *J. Fd. Sci.* 36: 786-790.
- Hornstein, I., P. F. Crowe and R. Hiner. 1968. Composition of lipids in some beef muscles. *J. Fd. Sci.* 32: 650-655.
- Howard, V. W. Jr. 1967. Identifying fecal groups by pH analysis. *J. Wildl. Manage.* 31: 190-191.
- Hsu, T. C., and K. Benirschke. 1969. An atlas of mammalian chromosomes. Vol. 3. Springer Verlag, New York.
- Hubbard, A. W., and W. D. Pocklington. 1968. Distribution of fatty acids in lipids as an aid to the identification of animal tissues. I.-Bovine, porcine, ovine and some avian species. *J. Sci. Fd. Agric.* 19: 571-577.
- Jones, P. F. 1975. New applications of photoluminescence techniques for forensic science. pp. 183-196. In: G. Davies (ed) *Forensic Science*. American Chemical Society, Washington, D.C.
- Keiss, R. W., and S. M. Morrison. 1956. Identification of Colorado big game animals by the precipitin reaction. *J. Wildl. Manage.* 20: 169-172.
- Kelsall, J. P., and R. Burton. 1977. Identification of origins of lesser snow geese by x-ray spectrometry. *Can. J. Zool.* 55: 718-732.
- _____, W. J. Pannekoek, and R. Burton. 1975. Variability in the chemical content of waterfowl plumage. *Can. J. Zool.* 53: 1379-1386.
- Kind, S. S., and M. Watson. 1973. The estimation of blood stain age from the spectrophotometric properties of ammoniacal blood stain extracts. *Forens. Sci.* 2: 325-332.
- Klecka, W. R. 1975. Discriminant analysis. pp. 434-467. In: Nie, N. H., C. H. Hull, J. G. Jenkins, K. Steinbrenner, and D. H. Blunt, eds. *SPSS: Statistical package for the social sciences*. 2nd. ed. McGraw-Hill Co., New York.

- Knight, R. R. 1966. Bone characteristics associated with aging in elk. *J. Wildl. Manage.* 30: 369-374.
- _____. 1969. Some chemical characteristics of elk blood. *Bull. Wildl. Dis. Assoc.* 5: 8-10.
- Krausman, P. R., E. D. Ables, and C. M. McGinnis. 1974. Deer identification through pellet pH. *J. Wildl. Manage.* 38: 572-573.
- Leat, W. M. F. 1970. Carbohydrate and lipid metabolism in the ruminant during post-natal development. pp. 211-222. In: *Physiology of digestion and metabolism in the ruminant.* A. T. Phillipson ed. Oriel Press Limited.
- Lee, L. D., K. Ludwig, and H. P. Baden. 1978. Matrix proteins of human hair as a tool for identification of individuals. *Forens. Sci.*, 11: 115-121.
- Love, R. M. 1962. Protein denaturation in frozen fish. VI-Cold-storage studies on cod using the cell fragility method. *J. Sci. Fd. Agric.* 13: 269-278.
- _____, and E. M. MacKay. 1962. Protein denaturation in frozen fish. V.-Development of the cell fragility method for measuring cold-storage changes in the muscle. *J. Sci. Fd. Agric.* 13: 200-212.
- Ludwig, J. and J. L. Titus. 1972. Chromosome study of autopsy tissues. pp. 217-219. In: *J. Ludwig, Current Methods of Autopsy Practice.* W. B. Saunders Co. Philadelphia, London, Toronto. 356 pp.
- Mackie, I. M. 1969. Identification of fish species by a modified polyacrylamide disc electrophoresis technique. *Assoc. Publ. Anal.* J. 7: 83-87.
- Marchello, J. A., M. Vavra, F. D. Dryden, and D. E. Ray. 1970. Influence of sex on certain constituents of bovine muscles. *J. Anim. Sci.* 31: 707-712.
- Markert, C. L., and F. Møller. 1959. Multiple forms of enzymes: tissue, ontogenetic, and species specific patterns. *Proc. Nat. Acad. Sci.* 45: 753-763.
- Miller, W. J., A. O. Haugen, and D. J. Roslien. 1965. Natural variation in the blood proteins of white-tailed deer. *J. Wildl. Manage.* 29: 717-723.
- Moore, D. H. 1945. Species differences in serum protein patterns. *J. Biol. Chem.* 161: 21-32.
- Moore, K. L. 1966. Sex chromatin patterns in various animals. pp. 27-30. In: *The sex chromatin.* K. L. Moore ed. W. B. Saunders Co., Philadelphia. 474 pp.

- Moore, T. D., L. E. Spence, and C. E. Dugnolle. 1974. Identification of the dorsal guard hairs of some mammals of Wyoming. Wyoming Game and Fish Dept. Bull. 14. Laramie, Wyo. 177 pp.
- Morse, W. B. 1971. Law enforcement - a tool of management, pp. 120-123. In: A Manual of Wildlife Conservation. R. D. Teague ed. The Wildlife Society, Washington, D.C.
- Nagy, J. G., and J. G. Gilbert. 1968. Fecal pH values of mule deer and grazing domestic sheep. J. Wildl. Manage. 32: 961-962.
- Oates, D. W., C. W. Brown, and D. L. Weigel. 1974. Blood and tissue identification of selected birds and mammals. Nebraska Game and Parks Commission. Lincoln, Nebr.
- _____, and D. L. Weigel. 1976. Blood and tissue identification of selected birds and mammals. Part II. Cross reaction patterns of birds and mammals, using deer, bovine, raccoon, chicken and mallard duck antiserum. Nebraska Game and Parks Commission. Lincoln, Nebr.
- O'Keefe, P. W., G. H. Wellington, L. R. Mattick, and J. R. Stouffer. 1968. Composition of bovine muscle lipids at various carcass locations. J. Fd. Sci. 33: 188-192.
- Payne, E. 1971. The use of the fatty acid composition of lipids in the identification of horse and kangaroo meat. J. Sci. Fd. Agric. 22: 520-522.
- _____. 1978. Fatty acid composition of tissue phospholipids of the foetal calf and neonatal lamb, deer calf and piglet as compared with the cow, sheep, deer and pig. Br. J. Nutr. 39: 45-52.
- Perkons, A. K., and R. E. Jarvis. 1962. Neutron activation applied to identification of biological and related materials of forensic importance. In: Proceedings of the Canadian Society of Forensic Science Vol. 1. (Abstract).
- Pothoven, M. A., D. C. Beitz, and A. Zimmerli. 1974. Fatty acid compositions of bovine adipose tissue and of in vitro lipogenesis. J. Nutr. 104: 430-433.
- Reichert, E. T., and A. P. Brown. 1909. The differentiation and specificity of corresponding proteins and other vital substances in relation to biological classification and organic evolution: the crystallography of hemoglobins. Carnegie Institute of Washington Publ. No. 116. Washington, D.C.
- Roberts, W. K. 1966. Effects of diet, degree of fatness, and sex upon fatty acid composition of cattle tissues. Can. J. Anim. Sci. 46: 181-190.

- Rumsey, T. S., R. R. Oltjen, K. P. Bovard, and B. M. Priode. 1972. Influence of widely diverse finishing regimens and breeding on depot fat composition in beef cattle. *J. Anim. Sci.* 35: 1069-1075.
- Schmid, E. 1972. Atlas of animal bones. Elsevier Publ. Co., Amsterdam. 159 pp.
- Shaw, J. C., W. L. Ensor, H. F. Tellechea, and S. D. Lee. 1960. Relation of diet to rumen volatile fatty acids, digestibility, efficiency of gain and degree of unsaturation of body fat in steers. *J. Nutr.* 71: 203-208.
- Short, H. L., D. E. Medin, and A. E. Anderson. 1966. Seasonal variations in volatile fatty acids in the rumen of mule deer. *J. Wildl. Manage.* 30: 466-470.
- Sibley, C. G., and P. A. Johnsgard. 1959. Variability in the electrophoretic patterns of avian serum proteins. *Condor* 61: 85-95.
- Sisson, S., and J. D. Grossman. 1953. The anatomy of the domestic animals. W. B. Saunders and Co., Philadelphia, Pa. 4th ed.
- Sturner, W. Q., and G. E. Gantner. 1964. The postmortem interval. A study of potassium in the vitreous humor. *Amer. J. Clin. Path.* 42: 137-144.
- Sumida, D. M., D. W. Vogt, E. H. Cobb, I. I. Iwanaga, and D. Reimer. 1972. Effect of breed type and feeding regime on fatty acid composition of certain bovine tissues. *J. Anim. Sci.* 35: 1058-1063.
- Sweet, G. H., and J. W. Elvins. 1976. Studies by crossed electroimmunodiffusion on the individuality and sexual origin of bloodstains. *J. Forens. Sci.* 21: 498-509.
- Taber, R. D. 1956. Characteristics of the pelvic girdle in relation to sex in black-tailed and white-tailed deer. *Calif. Fish and Game* 42: 15-21.
- Tanhuanpää, E., and E. Pulliainen. 1975. Major fatty acid composition of some organ fats in the moose (*Alces alces*) in northeastern Lapland. *Ann. Zool. Fennici* 12: 148-155.
- Tempelis, C. H., and M. L. Rodrick. 1973. Serologic technique for the identification of animal blood proteins. *J. Wildl. Manage.* 37: 584-585.
- Terrell, R. N., G. G. Suess, R. G. Cassens, and R. W. Bray. 1968. Broiling, sex and interrelationships with carcass and growth characteristics and their effect on the neutral and phospholipid fatty acids of the bovine longissimus dorsi. *J. Fd. Sci.* 33: 562-565.

- Tove, S. B. 1965. Fat metabolism in ruminants. In: 2nd International Symposium on the Physiology of Digestion in the Ruminant. pp. 399-410.
- Twibell, J. and P. H. Whitehead. 1978. Enzyme typing of human hair roots. J. Forens. Sci. 23: 356-360.
- Vakil, D. V., P. K. Lewin, and P. E. Conen. 1973. Value of fluorescent Y chromosomes and sex chromatin tests. Acta. Cytol. 17: 220-223.
- van Tets, P., and I. McT. Cowan. 1966. Some sources of variation in the blood sera of deer (*Odocoileus*) as revealed by starch gel electrophoresis. Can. J. Zool. 44: 631-647.
- Wagner, C. J. 1977. pp. 11-12 In: Forensic Science: Symposium Proceedings. Alberta Recreation, Parks and Wildlife.
- Washino, R. K., and J. G. Else. 1972. Identification of blood meals of hematophagous arthropods by the hemoglobin crystallization method. Amer. J. Trop. Med. Hyg. 21: 120-122.
- Werrett, D. J., L. A. King, and P. H. Whitehead. 1976. The detection of allergen-associated antibodies in bloodstains. J. Forens. Sci. Soc. 16: 121-126.
- Winter, K. B., and R. F. Honess. 1952. The use of transmission spectra and crystallography of hemoglobin in law enforcement. J. Wildl. Manage. 16: 111-113.
- Wishart, W. D. 1977. pp. 5-6. In: Forensic Science: Symposium Proceedings. Alberta Recreation, Parks and Wildlife.

APPENDIX I

Background data and fatty acid composition of muscle samples used in the present study. The identification number provides the following information: columns 1-3 = animal number; columns 4-6 = date sample was collected (Julian calendar); columns 7-8 = year sample was collected; column 9 = animal sex (1 = male, 2 = non-pregnant female, 3 = pregnant female, 4 = ? pregnant female); column 10-11 = animal age (01 = fetus, 04 = 4-6 months, 05 = 7-9 months, 06 = 10-12 months, 07 = 13-15 months, 08 = 16-24 months, 09 = >24 months); column 12 = portion of muscle (1 = proximal, 2 = mid, 3 = distal, 4 = whole); columns 13-14 = muscle (01 = l. dorsi, 02 = sterno-cephalicus, 03 = trapezius, 04 = infraspinatus, 05 = extensor carpi radialis, 06 = psoas, 07 = biceps femoris, 08 = semitendinosus, 09 = long digital extensor and peroneus tertius, 10 = semimembranosus, 11 = obliquus abdominis externus, 12 = supraspinatus).

		Fatty Acid											
Identification No.		14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Triacylglycerols													
Elk													
Adult Male													
	40006877108401	3.5	1.6	27.8	13.2	12.3	29.0	5.9	1.2	0.0	1.1	0.1	0.2
	40006877108404	4.4	1.8	27.4	9.4	16.4	28.0	5.1	1.6	0.0	0.3	0.0	0.0
	40006877108405	2.7	1.4	24.8	13.8	10.6	36.0	5.4	0.8	0.0	0.2	0.0	0.0
	40006877108407	4.3	1.6	30.1	9.8	16.5	25.8	5.4	1.0	0.0	1.2	0.1	0.2
	40006877108408	3.3	1.4	27.2	12.8	11.5	32.1	6.0	0.8	0.0	0.6	0.0	0.0
	40006877108409	2.9	1.3	25.5	13.2	12.2	34.7	4.9	0.6	0.0	0.2	0.0	0.0
	40302777109401	6.2	1.4	33.6	6.0	21.9	23.0	3.0	0.1	0.1	0.6	0.0	0.0
	40302777109404	3.9	1.6	25.5	7.8	20.9	29.1	4.2	0.9	0.1	0.3	0.0	0.0
	40302777109405	2.1	1.0	24.5	10.1	16.6	36.3	4.4	0.5	0.0	0.3	0.0	0.0
	40302777109407	3.3	1.0	31.0	8.4	19.0	25.7	6.1	0.5	0.1	1.1	0.0	0.0
	40302777109408	4.5	1.4	30.2	7.2	19.3	27.6	3.8	0.6	0.1	0.3	0.0	0.0
	40302777109409	3.7	1.7	26.5	12.2	15.3	31.9	3.3	0.5	0.0	0.2	0.0	0.0
	77003174109301	8.2	2.4	43.7	12.8	8.2	20.0	2.2	0.4	0.0	0.0	0.0	0.0
	78003574109301	11.6	3.4	42.8	12.1	8.7	17.6	1.5	0.3	0.0	0.0	0.0	0.0
	93603574108301	8.5	3.6	38.0	16.9	7.4	19.5	2.2	0.6	0.0	0.0	0.0	0.0
Adult Female													
	40109077209401	4.9	2.0	32.2	14.6	12.0	25.6	3.6	0.6	0.0	0.2	0.0	0.0
	40109077209404	3.4	1.5	25.4	12.2	14.1	32.6	5.1	1.3	0.0	0.3	0.0	0.0
	40109077209405	3.1	2.2	24.1	16.2	9.8	36.2	3.4	0.8	0.1	0.2	0.0	0.0
	40109077209407	3.9	1.9	27.9	14.1	11.5	31.1	3.7	0.9	0.0	0.3	0.0	0.1
	40109077209408	4.2	2.5	27.7	17.0	9.1	31.2	3.3	0.8	0.0	0.4	0.0	0.0
	40109077209409	3.6	2.8	22.9	18.0	9.6	34.1	3.2	1.0	0.0	0.2	0.0	0.0
	40200377409401	6.0	3.1	33.1	15.7	8.4	27.4	2.0	0.5	0.0	0.3	0.0	0.0
	40200377409404	4.1	2.2	27.5	15.2	9.6	33.2	3.2	1.1	0.0	0.2	0.0	0.0
												. . . Continued	

APPENDIX I (Continued).

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Triacylglycerols												
Elk												
Adult Female												
40200377409405	2.9	2.5	23.4	21.5	4.3	39.3	2.4	0.6	0.1	0.2	0.0	0.0
40200377409407	4.2	2.1	30.3	17.2	7.1	33.1	2.3	0.6	0.0	0.6	0.0	0.0
40200377409408	3.9	2.6	27.6	18.5	6.2	34.2	2.6	0.7	0.1	0.3	0.0	0.0
40200377409409	2.7	1.9	23.7	20.9	5.7	39.8	2.0	0.5	0.1	0.2	0.0	0.0
40436276409401	5.8	3.9	29.3	20.6	5.5	25.0	3.7	0.3	0.0	0.1	0.0	0.0
40436276409404	4.7	2.7	26.0	16.6	9.0	31.0	4.7	0.4	0.0	0.1	0.0	0.0
40436276409405	3.6	3.2	22.4	20.2	6.3	36.4	3.9	0.6	0.0	0.5	0.0	0.0
40436276409407	6.2	4.1	30.8	20.8	6.0	27.0	2.2	0.4	0.0	0.0	0.0	0.0
40436276409408	4.4	3.2	28.1	23.4	4.4	30.3	3.1	0.2	0.0	0.1	0.0	0.0
40436276409409	3.9	4.6	20.5	25.1	3.4	33.0	3.5	0.2	0.0	0.1	0.0	0.0
52103174309301	8.3	4.4	35.6	18.1	5.4	22.2	2.5	0.6	0.0	0.0	0.0	0.0
77703274208301	6.6	3.3	38.2	20.1	5.8	22.3	1.4	0.4	0.0	0.0	0.0	0.0
77803274309301	5.2	2.8	34.3	17.5	7.0	28.4	2.1	0.5	0.0	0.0	0.0	0.0
78203574309301	5.7	2.2	39.2	17.6	6.5	24.5	1.8	0.3	0.0	0.0	0.0	0.0
92603274309301	6.8	3.6	35.3	17.1	6.4	24.0	2.5	0.8	0.0	0.0	0.0	0.0
92703274309301	9.4	5.0	35.7	16.8	6.6	21.4	2.0	0.5	0.0	0.0	0.0	0.0
94103674309301	8.4	3.2	41.5	16.0	6.9	20.5	1.2	0.2	0.0	0.0	0.0	0.0
40813877309405	3.1	2.0	25.2	16.1	7.0	40.0	2.8	0.7	0.0	0.6	0.0	0.0
40813877309404	2.9	2.0	24.7	16.7	6.7	40.1	3.0	0.7	0.0	0.5	0.0	0.0
40813877309401	4.7	2.0	31.2	13.9	10.0	26.5	5.9	1.2	0.0	1.0	0.0	0.0
40813877309409	3.6	2.3	23.7	15.7	8.2	38.5	3.4	0.9	0.0	0.7	0.0	0.0
Juvenile Male												
75703074105301	2.8	0.3	25.9	2.7	27.6	27.5	4.0	0.4	0.0	0.0	0.0	0.0
40736176105405	4.7	2.4	23.1	12.8	11.8	33.9	5.3	0.9	0.1	0.3	0.0	0.0
. . . Continued												

APPENDIX I (Continued)

Identification No.		14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Triacylglycerols													
Elk													
Juvenile Male													
	40736176105404	5.2	1.8	24.8	9.1	14.9	31.9	5.4	0.5	0.0	0.1	0.0	0.0
	40736176105407	4.8	1.7	28.0	10.4	14.8	31.5	3.6	0.5	0.0	0.1	0.0	0.0
	40736176105408	3.3	1.2	25.0	9.3	15.7	35.6	4.2	0.6	0.0	0.1	0.0	0.0
	40736176105409	4.0	1.9	23.1	11.2	13.4	31.5	5.8	0.7	0.1	0.9	0.0	0.0
Juvenile Female													
	75903074205301	5.8	0.6	40.0	4.0	19.2	21.4	5.5	0.4	0.0	0.0	0.0	0.0
Fetal Male													
	04402874101301	3.1	0.1	39.9	5.8	17.7	23.8	2.5	0.1	0.4	1.6	0.0	0.6
Fetal Female													
	001 76201301	3.9	0.5	45.4	6.8	13.6	22.5	2.7	0.3	0.0	1.9	0.0	0.0
Moose													
Adult Male													
	00202874109301	0.9	0.4	18.2	4.9	11.7	56.2	2.1	1.0	0.0	0.0	0.0	0.0
	00302874109301	0.5	0.3	16.4	1.8	29.2	42.9	3.3	0.6	0.0	0.0	0.0	0.0
	21825376109301	1.8	0.8	17.4	2.1	21.8	46.9	2.5	1.1	0.0	0.1	0.0	0.0
	40525077108301	0.7	0.2	21.6	5.0	16.2	48.2	3.2	0.3	0.1	0.3	0.0	0.0
	40602577109401	0.7	0.2	15.7	2.6	23.2	46.5	4.5	1.0	0.0	0.0	0.0	0.0
	40602577109404	0.7	0.3	16.9	2.8	20.9	45.7	5.2	1.6	0.1	0.2	0.0	0.0
	40602577109405	0.6	0.2	16.0	2.9	14.9	53.4	5.1	1.3	0.0	0.2	0.0	0.0
	40602577109407	0.6	0.2	13.8	2.2	22.0	50.2	4.4	1.0	0.0	0.2	0.0	0.0
											. . . Continued		

APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Triacylglycerols												
Moose												
Adult Male												
40602577109408	0.3	0.1	13.8	2.2	20.8	49.9	6.1	1.2	0.1	0.3	0.0	0.0
40602577109409	0.7	0.2	15.5	3.0	16.0	54.5	4.0	1.0	0.0	0.2	0.0	0.0
51803274109301	1.3	0.5	20.0	2.1	24.5	42.8	3.4	0.5	0.0	0.0	0.0	0.0
75102974109301	0.6	0.2	16.6	1.9	28.1	41.8	3.6	0.7	0.0	0.0	0.0	0.0
91503174108301	2.0	0.3	21.6	2.2	27.3	36.4	4.1	1.4	0.0	0.0	0.0	0.0
92303274109301	0.6	0.2	15.5	1.6	28.8	43.4	3.6	1.0	0.0	0.0	0.0	0.0
42028075109102	0.2	0.0	7.8	1.0	35.3	46.9	4.1	1.0	0.0	0.0	0.0	0.0
41708878109301	0.7	0.2	16.4	2.1	22.9	44.2	4.4	1.7	0.0	0.2	0.0	0.0
Adult Female												
02803174309301	1.3	0.4	23.2	4.1	19.5	43.1	5.2	1.3	0.0	0.0	0.0	0.0
22434675209301	1.3	0.0	21.6	1.5	33.6	34.5	3.9	0.6	0.0	0.0	0.0	0.0
52703574309301	0.8	0.1	17.2	1.9	25.6	48.4	2.0	0.7	0.0	0.0	0.0	0.0
53003574309301	1.0	0.2	19.8	2.0	22.1	45.8	4.2	1.1	0.0	0.0	0.0	0.0
74302874309301	0.9	0.2	19.7	2.1	23.6	45.2	3.0	1.0	0.0	0.0	0.0	0.0
74902974309301	0.7	0.2	22.6	2.0	24.3	41.3	3.1	0.9	0.0	0.0	0.0	0.0
78403574208301	1.2	0.2	17.1	1.8	24.8	45.6	3.6	0.9	0.0	0.0	0.0	0.0
90702974309301	1.0	0.2	21.4	2.0	26.5	41.0	2.3	0.6	0.0	0.0	0.0	0.0
93003274309301	1.5	0.2	19.1	2.7	20.7	48.7	3.6	1.6	0.0	0.0	0.0	0.0
22434675209305	2.5	0.7	26.1	8.4	17.8	31.9	5.7	0.6	0.0	0.0	0.0	0.0
22434675209314	1.1	0.6	14.3	1.1	27.6	34.9	5.6	0.7	0.0	0.0	0.0	0.0
22434675209303	1.2	0.3	17.7	1.4	33.9	30.3	8.3	1.0	0.0	0.0	0.0	0.0
22434675209306	1.4	0.3	15.2	1.1	32.5	29.3	9.4	1.2	0.0	0.0	0.0	0.0
22434675209307	1.5	0.3	15.1	1.2	35.1	34.4	6.4	0.9	0.0	0.0	0.0	0.0
22434675209309	2.0	1.0	21.6	6.1	19.7	26.7	13.4	1.3	0.0	0.0	0.0	0.0
22434675209302	0.9	0.5	14.1	1.2	42.0	31.2	4.3	0.8	0.0	0.0	0.0	0.0
41809078309301	1.1	0.2	21.1	2.4	26.6	41.0	3.6	0.6	0.0	0.0	0.0	0.0

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APPENDIX I (Continued)

Triacylglycerols		Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Moose														
Juvenile Male														
		77303274105301	1.9	0.5	20.3	2.8	25.1	37.3	4.3	1.5	0.0	0.0	0.0	0.0
Juvenile Female														
		93203574205301	2.6	0.8	23.5	2.6	24.9	31.0	3.4	0.7	0.0	0.0	0.0	0.0
		40913877206405	0.7	0.5	24.0	4.2	20.1	29.9	8.5	1.4	0.2	1.8	0.0	0.0
		40913877206404	1.3	1.1	24.1	5.4	20.9	30.5	4.3	1.0	0.0	0.2	0.0	0.0
		40913877206407	1.4	1.2	23.7	5.5	20.1	31.6	4.7	0.9	0.0	0.5	0.0	0.0
		40913877206408	1.0	0.4	24.6	5.3	20.0	32.5	4.5	0.8	0.0	0.4	0.0	0.0
		40913877206409	0.9	0.6	24.0	5.0	17.9	34.2	6.5	1.1	0.2	1.3	0.0	0.0
		41100477205405	1.4	0.4	20.0	4.0	20.1	41.0	4.7	1.2	0.1	0.1	0.0	0.0
		41100477205404	1.8	0.6	20.4	3.9	22.0	34.8	7.0	1.5	0.1	0.2	0.0	0.0
		41100477205401	2.2	0.4	21.4	2.8	27.2	33.2	5.4	1.6	0.1	0.3	0.0	0.0
		41100477205409	1.8	0.6	22.0	4.8	19.2	36.8	6.7	1.2	0.2	0.0	0.0	0.0
Fetal Male														
		01202874101301	3.0	0.0	37.6	9.5	14.3	28.3	2.4	0.4	0.5	0.6	0.0	0.0
		04902974101301	3.8	0.0	40.0	9.0	13.4	25.4	2.2	0.1	1.0	0.9	0.1	0.0
Fetal Female														
		00602974201301	3.2	0.0	40.4	8.5	14.0	27.5	1.6	0.1	0.6	0.7	0.1	0.0
		00702974201301	3.2	0.2	39.4	9.6	15.2	26.2	2.1	0.2	0.5	0.4	0.1	0.0
		01302974201301	2.6	0.0	37.0	10.1	13.3	27.9	2.1	0.8	0.7	0.6	0.2	0.0
White-Tailed Deer														
Adult Male														
		02633474108301	0.3	0.1	18.2	2.0	29.6	43.4	1.9	0.8	0.0	0.0	0.0	0.0
		99603274109301	1.4	0.2	24.8	1.4	31.9	33.0	2.6	0.7	0.0	0.0	0.0	0.0
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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Triacylglycerols												
White-Tailed Deer												
Adult Male												
99803274108301	1.2	0.0	20.1	1.7	34.5	36.8	2.6	0.2	0.0	0.0	0.0	0.0
31232575109211	1.5	0.2	23.7	0.5	29.2	40.2	1.5	1.0	0.0	0.1	0.0	0.0
31332675109211	1.3	0.1	21.5	0.9	26.7	41.0	2.9	1.8	0.0	0.1	0.0	0.0
31632575109211	1.1	0.1	20.3	0.6	30.9	40.5	1.7	1.3	0.0	0.1	0.0	0.0
31832575109211	1.3	0.1	23.6	1.3	18.8	46.6	1.5	0.7	0.0	0.1	0.0	0.0
31932675109211	1.3	0.1	23.8	0.5	27.5	41.3	2.3	0.9	0.0	0.1	0.0	0.0
32032575108211	0.9	0.0	23.8	1.4	31.1	37.2	2.1	1.3	0.0	0.1	0.0	0.0
32332575109211	1.5	0.1	21.5	1.6	27.1	42.0	1.9	1.3	0.0	0.0	0.0	0.0
41218877109405	1.7	0.4	21.6	4.7	17.0	41.3	6.7	2.0	0.0	0.6	0.1	0.0
41218877109404	1.3	0.3	19.8	3.5	21.6	29.0	13.4	4.1	0.2	2.6	0.6	0.0
41218877109401	1.5	0.5	21.3	3.8	25.8	27.6	9.4	1.4	0.1	4.4	0.2	0.0
41218877109407	0.7	0.2	20.1	3.0	26.9	35.3	7.1	1.8	0.1	1.1	0.2	0.0
41218877109408	0.9	0.3	18.6	3.5	22.7	39.1	7.5	2.3	0.1	0.8	0.2	0.0
41218877109409	2.0	0.5	27.1	3.0	27.0	29.2	4.7	1.0	0.0	0.9	0.0	0.0
41502377108405	2.0	0.5	24.6	4.2	18.6	38.1	5.5	0.9	0.0	0.1	0.0	0.0
41502377108404	2.6	0.7	25.6	3.0	25.8	30.5	4.7	1.6	0.1	0.3	0.1	0.0
41502377108408	1.5	0.4	21.4	3.4	22.6	38.6	6.3	0.9	0.0	0.3	0.0	0.0
41502377108409	1.9	0.6	20.9	4.6	16.9	44.2	6.0	0.8	0.0	0.1	0.0	0.0
Adult Female												
22902775309301	0.6	0.1	20.6	2.2	23.7	46.7	2.4	1.0	0.0	0.0	0.0	0.0
33203274209301	0.8	0.2	17.9	1.4	31.2	41.3	2.7	0.8	0.0	0.0	0.0	0.0
33303274309301	0.7	0.0	21.7	1.7	28.4	41.1	3.0	0.6	0.0	0.0	0.0	0.0
34003274309301	0.9	0.1	19.5	1.6	28.8	43.5	2.5	0.7	0.0	0.0	0.0	0.0
91703174308301	1.0	0.2	18.2	2.1	22.6	48.9	2.9	0.8	0.0	0.0	0.0	0.0
99903274309301	1.1	0.3	22.3	1.8	30.1	37.4	3.0	1.0	0.0	0.0	0.0	0.0

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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Triacylglycerols												
Mule Deer												
Adult Female												
22332575209301	1.3	0.2	20.0	2.3	26.7	39.8	2.9	0.9	0.0	0.3	0.0	0.1
22818175209301	0.4	0.1	16.8	1.7	32.7	38.0	3.9	2.7	0.0	0.0	0.0	0.0
30333075209211	2.2	0.1	28.7	1.0	22.0	41.7	1.5	1.1	0.0	0.1	0.0	0.0
30433275209211	1.8	0.1	28.1	0.6	31.7	33.6	1.4	0.7	0.0	0.0	0.0	0.0
Juvenile Male												
22719975107301	0.9	0.5	18.5	2.1	34.7	32.7	3.5	2.9	0.0	0.0	0.0	0.0
Immature Male												
22033075104301	4.4	1.0	26.0	3.9	18.5	37.3	2.6	1.2	0.0	0.1	0.0	0.0
30132575104211	2.0	0.1	29.0	0.8	25.3	37.0	1.9	1.2	0.0	0.1	0.0	0.0
30232575104211	0.5	0.1	21.2	0.8	30.1	41.8	1.9	1.1	0.0	0.1	0.0	0.0
30732975104211	1.9	0.2	24.9	0.7	27.5	37.9	3.3	1.0	0.0	0.3	0.0	0.0
Immature Female												
30933875204301	2.0	0.1	28.9	1.2	23.1	38.7	2.4	0.9	0.0	0.0	0.0	0.0
Antelope												
Adult Male												
20130075109301	4.9	1.3	17.8	2.5	24.6	35.9	2.8	0.8	0.3	0.4	0.1	0.0
20230075109301	4.1	0.9	20.4	2.9	29.1	28.4	3.0	0.8	0.2	0.2	0.2	0.0
20325376108301	1.8	0.5	18.9	2.5	28.9	38.3	2.8	0.4	0.0	0.3	0.1	0.0
20430076109301	4.0	1.8	19.1	2.6	23.1	36.2	3.2	0.7	0.0	0.6	0.1	0.0
20530076109301	3.5	0.7	26.5	2.9	22.1	36.5	2.6	1.3	0.0	0.3	0.1	0.0
23430076109401	1.1	0.4	15.9	3.3	23.0	45.1	5.2	0.4	0.1	0.3	0.0	0.0

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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Triacylglycerols												
Antelope												
Adult Male												
23430076109404	1.6	0.5	20.2	2.4	28.9	37.8	2.3	0.7	0.0	0.1	0.0	0.0
23430076109405	1.3	0.5	17.8	4.0	18.0	48.9	4.5	0.3	0.0	0.4	0.0	0.0
23430076109407	1.5	0.4	19.8	2.7	26.2	42.1	1.5	0.3	0.0	0.0	0.0	0.0
23430076109408	1.1	0.4	18.2	2.7	24.1	43.8	2.6	0.5	0.0	0.5	0.2	0.0
23430076109409	1.5	0.6	16.9	4.5	18.4	47.6	4.1	0.3	0.2	0.0	0.0	0.0
23430076109410	0.7	0.3	19.1	2.3	27.5	43.5	1.7	0.4	0.0	0.2	0.0	0.0
Bighorn Sheep												
Adult Female												
20628076209301	3.7	0.8	22.2	3.6	15.7	44.4	3.2	1.6	0.0	0.1	0.1	0.0
20728076209301	3.6	0.6	23.2	2.4	20.2	43.7	2.2	1.0	0.0	0.1	0.0	0.0
20827976209301	4.2	0.8	22.9	3.4	18.0	41.9	2.8	1.3	0.0	0.2	0.1	0.0
20928076209301	4.6	1.3	22.3	3.4	16.3	43.3	2.6	1.4	0.0	0.2	0.0	0.0
21027976209206	4.6	1.1	22.2	3.5	17.6	41.5	2.8	1.6	0.0	0.1	0.0	0.0
21126676209301	3.7	0.6	24.5	3.6	16.2	43.4	2.4	1.2	0.0	0.1	0.1	0.0
21226576209301	3.3	0.5	23.3	3.4	16.0	45.8	2.4	1.3	0.0	0.1	0.0	0.0
21627976209206	3.7	0.4	25.5	2.9	16.1	44.5	3.2	1.3	0.0	0.5	0.0	0.0
23507377409401	2.0	0.4	20.1	4.7	18.4	43.2	5.8	1.2	0.0	0.1	0.0	0.0
23507377409407	2.6	0.5	20.7	5.1	16.0	44.8	5.5	1.2	0.0	0.0	0.0	0.0
23507377409408	2.5	0.5	20.6	5.6	14.7	47.2	4.3	0.6	0.0	0.0	0.0	0.0
23507377409409	1.9	0.4	19.4	5.3	11.8	47.4	7.6	1.2	0.0	1.2	0.5	0.0

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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
Elk												
Adult Male												
40006877108401	1.0	0.0	9.2	3.3	13.7	12.9	31.0	4.0	1.3	12.6	3.6	3.6
40006877108404	0.2	0.0	12.4	2.6	15.2	12.6	28.6	5.2	1.1	12.1	1.9	3.0
40006877108405	0.2	0.0	10.7	3.0	14.6	10.5	26.7	4.3	0.8	11.8	3.3	3.5
40006877108407	0.3	0.0	14.5	2.3	12.1	10.8	29.0	3.8	1.4	13.4	3.4	4.3
40006877108408	0.1	0.0	9.0	3.1	12.9	10.9	28.0	4.5	0.9	12.0	3.5	3.6
40006877108409	0.2	0.0	9.6	2.8	15.7	10.6	27.3	4.8	0.7	12.2	3.1	3.3
40302777109401	0.2	0.1	12.3	2.1	12.9	12.3	27.3	3.8	1.9	17.1	3.9	3.7
40302777109404	0.1	0.2	11.2	1.6	16.8	12.4	29.2	3.6	1.8	14.9	1.8	2.4
40302777109405	0.2	0.3	17.3	2.3	14.5	10.9	23.1	4.1	1.5	15.8	3.3	3.3
40302777109407	0.2	0.0	19.9	2.4	18.7	12.8	24.7	3.1	1.0	8.3	1.1	1.0
40302777109408	0.2	0.4	21.9	2.5	12.8	10.4	22.3	3.0	1.7	12.1	2.7	2.7
40302777109409	0.3	0.2	11.8	1.6	16.3	11.6	26.0	4.4	1.6	17.0	3.3	3.8
77003174109301	0.4	0.2	12.7	2.9	15.4	15.4	29.8	2.8	2.2	12.0	1.7	2.5
78003574109301	0.4	0.2	21.4	2.8	19.7	11.8	25.8	1.2	1.6	10.4	1.5	0.8
93603574108301	0.5	0.2	11.9	2.7	18.1	14.8	31.8	3.5	1.3	9.3	2.0	2.3
Adult Female												
40109077209401	0.3	0.2	9.3	3.6	13.8	11.7	28.3	3.0	1.3	13.8	3.0	3.0
40109077209404	0.2	0.2	9.4	3.7	13.7	13.0	30.3	3.5	2.2	14.3	2.6	3.3
40109077209405	0.3	0.2	16.8	3.6	17.2	11.5	23.6	2.5	1.0	13.8	2.3	2.7
40109077209407	0.3	0.2	13.6	3.2	15.0	10.9	27.0	2.6	1.5	17.1	2.7	2.6
40109077209408	0.3	0.4	14.9	3.4	14.7	9.8	27.3	2.5	1.5	14.7	4.9	2.7
40109077209409	0.2	0.2	8.2	2.3	15.1	9.1	25.8	2.9	1.9	18.7	4.6	3.6
40200377409401	0.6	0.6	17.7	3.7	23.9	13.2	23.8	1.7	0.9	7.5	1.7	1.2
40200377409404	0.5	0.4	16.5	5.1	16.8	14.5	23.9	3.1	0.9	10.6	1.7	2.0
40200377409405	0.2	0.2	19.1	3.4	18.6	9.0	24.8	3.3	1.0	12.1	2.8	2.4
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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
Elk												
Adult Female												
40200377409407	0.3	0.3	15.3	3.2	19.6	10.7	23.9	2.7	1.2	13.9	3.0	2.1
40200377409408	0.3	0.3	18.6	3.6	20.2	10.3	23.2	2.1	1.0	12.6	2.6	2.1
40200377409409	0.2	0.2	12.0	2.9	19.6	11.1	26.3	3.6	1.2	14.4	3.1	2.5
40436276409401	0.4	0.2	15.2	6.1	12.7	14.4	25.6	3.0	1.2	12.6	2.4	3.3
40436276409404	0.5	0.2	20.3	8.1	14.3	16.5	22.2	2.2	0.7	8.9	0.7	2.0
40436276409405	0.2	0.3	20.3	5.6	16.9	13.5	21.7	1.9	1.4	10.7	1.9	2.5
40436276409407	0.6	0.3	18.8	7.1	14.1	14.1	20.4	2.0	1.3	11.4	1.8	2.3
40436276409408	0.4	0.2	19.5	6.3	14.9	14.6	23.3	2.1	1.2	10.2	1.6	2.2
40436276409409	0.4	0.3	17.0	6.4	14.0	14.5	22.5	2.8	0.9	12.7	2.2	3.2
40813877309405	0.1	0.1	13.8	2.5	12.9	19.2	20.4	3.8	1.3	12.3	2.2	2.7
40813877309404	0.2	0.0	19.2	3.5	14.2	13.7	22.7	3.2	1.2	10.1	2.3	2.0
40813877309401	1.0	0.5	20.1	5.6	12.9	16.1	21.0	2.6	1.4	10.8	1.8	2.1
40813877309407	0.2	0.5	15.1	3.3	15.9	11.6	27.5	3.8	1.8	12.1	2.2	2.4
40813877309409	0.1	0.0	12.6	1.2	19.9	7.8	29.0	6.2	1.0	12.1	3.4	3.6
52103174309301	0.7	0.4	15.2	3.9	15.7	14.6	30.5	3.4	1.2	10.5	2.6	2.6
77703274208301	0.4	0.0	17.5	2.4	15.4	12.4	31.0	2.6	1.1	9.8	3.2	2.8
77803274309301	0.9	0.4	26.9	5.4	15.6	14.7	24.4	2.0	0.4	6.4	1.2	1.7
92603274309301	0.5	0.2	20.5	4.0	14.8	12.4	30.8	3.1	1.3	8.9	2.6	2.4
92703274309301	1.1	0.5	30.2	6.2	13.5	14.5	20.7	2.0	1.0	7.0	2.0	2.6
Juvenile Male												
40736176105405	0.4	0.0	11.1	1.9	18.0	14.1	30.6	3.1	1.6	10.7	2.0	2.5
40736176105404	0.4	0.4	15.0	3.9	16.2	15.4	26.9	3.6	0.9	9.8	1.4	2.2
40736176105407	0.4	0.5	15.7	3.1	15.0	11.9	28.8	3.0	1.4	9.8	2.1	2.1
40736176105408	0.3	0.3	11.8	2.2	17.1	12.8	33.0	3.0	1.4	10.1	2.2	2.4
40736176105409	0.5	0.5	16.4	3.1	17.6	12.6	26.1	2.8	1.2	10.0	1.7	2.3
75703074105301	0.3	0.1	8.6	1.1	14.7	10.2	36.2	2.6	1.6	13.4	5.7	3.8

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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
Elk												
Juvenile Female												
75903074205301	0.5	0.0	12.7	1.5	16.0	9.8	35.6	3.1	1.4	10.4	3.9	2.8
Fetal Male												
04402874101301	0.8	0.0	28.4	2.7	19.8	29.2	2.4	0.1	0.9	7.0	0.2	2.7
Moose												
Adult Male												
21825376109301	2.6	0.7	16.6	2.6	14.0	19.6	17.3	4.2	0.7	7.6	2.0	1.8
40525077108301	0.1	0.1	13.6	2.2	13.4	21.9	25.4	1.8	1.2	12.4	1.0	1.5
40602577109401	0.1	0.1	16.0	2.4	16.5	13.8	27.7	3.3	0.9	10.8	2.4	1.6
40602577109404	0.1	0.1	10.0	1.4	14.8	15.7	31.9	2.9	1.3	12.2	2.0	2.5
40602577109405	0.1	0.2	14.2	2.2	19.6	12.2	28.7	2.8	0.9	10.6	1.9	2.4
40602577109407	0.1	0.2	15.6	2.6	17.8	12.1	26.4	2.8	1.1	11.7	2.4	2.0
40602577109408	0.1	0.1	14.2	2.3	18.8	11.3	29.1	2.5	1.0	11.7	2.3	2.1
40602577109409	0.1	0.1	11.8	1.9	20.3	11.8	28.8	2.6	1.0	12.2	2.5	2.6
51803274109301	0.2	0.0	12.6	1.1	14.6	10.2	37.8	2.6	0.9	12.1	2.0	2.2
91503174108301	0.3	0.0	14.2	1.3	15.0	13.5	39.0	3.7	0.4	7.2	1.8	1.0
92303274109301	0.3	0.2	14.7	1.6	14.7	14.7	34.8	2.7	0.8	9.5	2.1	1.0
41708878109301	0.1	0.0	10.4	1.0	14.1	16.3	32.7	3.1	1.2	10.6	2.4	1.0
Adult Female												
02803274309301	0.9	0.5	19.4	3.0	16.5	17.3	29.1	2.2	0.4	6.7	1.4	2.7
03803274309301	0.6	0.1	16.1	1.7	15.7	19.2	31.8	3.2	0.4	8.1	2.1	2.4
22434675209305	0.1	0.0	9.9	0.9	18.4	13.8	34.8	3.7	0.4	12.4	2.3	3.2
22434675209314	0.0	0.2	16.1	1.9	18.7	10.2	31.0	3.5	0.4	11.4	2.6	2.7
22434675209303	0.0	0.0	16.0	1.6	21.3	14.1	30.6	2.7	0.3	8.5	1.6	2.2
. . . Continued												

APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
Moose												
Adult Female												
22434675209306	0.0	0.0	12.8	1.4	16.7	10.0	35.9	3.7	0.5	12.1	2.9	2.7
22434675209301	0.2	0.0	12.0	0.9	13.7	13.2	36.2	4.0	0.6	11.0	2.9	2.6
22434675209307	0.1	0.0	11.8	1.0	16.0	11.0	37.9	4.0	0.5	11.8	2.5	2.3
22434675209309	0.3	0.0	11.4	0.9	17.4	11.0	34.2	4.3	0.5	11.9	3.1	3.1
22434675209302	0.1	0.0	9.0	1.0	15.4	13.6	37.2	3.5	0.6	12.3	3.2	2.6
52703574309301	0.2	0.1	23.1	2.0	17.0	18.0	23.6	1.8	0.6	7.4	1.9	1.3
53003574309301	0.4	0.0	24.3	1.9	20.9	15.7	26.1	1.4	0.4	4.4	1.0	0.6
78403574208301	0.3	0.0	18.9	2.3	21.2	15.5	26.3	2.6	0.5	5.9	1.3	0.8
41809078309301	0.3	0.0	16.5	1.3	22.6	13.8	27.5	0.8	0.8	8.4	1.2	0.8
Juvenile Male												
77303274105301	0.4	0.0	16.8	1.5	15.9	16.5	32.0	3.0	0.6	6.5	1.5	1.4
Juvenile Female												
40913877206405	0.2	0.1	16.0	3.2	14.8	21.7	19.7	3.4	0.9	9.8	1.6	2.0
40913877206404	0.1	0.2	13.5	2.3	13.3	18.0	22.4	3.8	1.3	11.2	2.3	2.6
40913877206407	0.1	0.0	15.4	3.7	13.6	16.4	20.9	3.6	1.3	12.7	2.0	2.1
40913877206408	0.1	0.3	12.6	4.0	12.1	16.4	19.0	3.1	1.1	9.7	1.7	2.0
40913877206409	0.1	0.2	13.3	2.3	14.5	19.0	21.3	3.5	1.1	12.0	1.9	2.5
93203574205401	0.6	0.0	20.9	2.1	19.3	16.5	26.0	2.3	0.4	5.4	0.9	0.5
41100477205405	0.1	0.0	9.9	1.5	12.6	14.8	34.6	5.6	0.8	10.8	3.0	3.2
41100477205404	0.1	0.0	13.2	1.1	16.4	14.5	33.1	4.0	0.5	9.1	1.9	2.0
41100477205401	0.1	0.0	11.8	1.3	13.9	12.1	31.6	3.4	1.0	11.5	3.0	2.1
41100477205409	0.2	0.0	15.4	1.7	25.6	14.3	25.9	1.9	0.4	6.0	1.3	1.2
Fetal Male												
01202874101301	1.0	0.0	25.1	3.2	20.1	30.7	2.5	0.2	1.0	5.9	0.3	2.4
04902974101301	0.8	0.0	23.4	3.2	19.2	28.2	2.0	0.2	1.5	6.6	0.9	4.2

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APPENDIX I (Continued)

Phospholipids	Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Moose													
Fetal Female													
	00602974201301	0.7	0.0	27.4	2.5	21.9	32.8	1.3	0.1	0.9	5.1	0.5	1.5
	00702974201301	1.0	0.0	24.7	3.5	19.1	31.2	2.1	0.2	1.1	7.0	0.3	3.0
	01302974201301	0.8	0.0	23.2	2.8	18.7	34.8	1.5	0.1	1.3	6.2	0.3	2.5
White-Tailed Deer													
Adult Male													
	31232575109211	0.1	0.0	19.4	2.2	15.5	26.3	17.3	3.2	1.0	7.0	1.4	2.3
	31332675109211	0.1	0.0	16.4	2.0	17.5	16.8	28.0	3.8	0.7	7.2	1.9	2.8
	31632575109211	0.1	0.0	16.9	2.1	18.0	21.4	21.6	3.5	0.7	9.6	1.5	2.2
	31832575109211	0.0	0.0	16.2	2.1	16.3	27.6	22.4	3.1	0.6	6.8	1.1	1.6
	31932675109211	0.2	0.0	16.4	1.5	14.4	21.2	25.9	3.4	0.9	9.2	2.2	3.4
	32332575109211	0.3	0.2	21.1	2.0	18.2	18.8	18.9	3.5	0.7	7.8	1.6	2.4
	41218877109405	0.1	0.0	12.3	1.6	17.8	9.4	28.7	7.0	0.8	11.4	3.0	4.1
	41218877109404	0.2	0.1	14.3	1.6	19.5	11.7	28.1	5.4	0.7	8.3	3.6	2.5
	41218877109401	0.3	0.0	16.9	3.1	20.1	10.1	25.8	5.4	0.6	9.4	2.3	2.3
	41218877109407	0.1	0.0	14.1	2.1	17.7	9.6	28.4	6.0	1.0	10.6	2.8	3.4
	41218877109408	0.1	0.1	13.3	1.7	19.6	9.8	28.3	6.2	0.8	9.7	2.7	3.5
	41218877109409	0.1	0.0	11.0	1.3	19.0	8.4	29.7	7.0	0.8	11.6	3.1	3.9
	32032575108211	0.1	0.0	20.4	1.1	16.0	16.3	22.2	3.7	0.6	7.6	1.8	2.8
	41502377108405	0.1	0.0	14.1	2.7	16.8	12.3	28.3	3.4	0.8	12.8	2.9	3.7
	41502377108404	0.6	0.1	19.3	3.1	22.8	15.0	24.4	2.1	0.6	5.6	0.9	1.5
	41502377108408	0.1	0.0	13.2	2.5	17.0	12.8	29.1	3.4	0.9	12.9	2.8	3.4
	41502377108409	0.1	0.0	11.5	2.5	18.0	13.8	27.0	3.5	0.8	13.7	2.8	3.6
	99603274109301	0.2	0.0	15.6	1.5	16.2	13.1	30.9	4.9	0.6	10.2	2.6	2.2
	99803274108301	0.3	0.2	20.1	2.3	17.0	11.4	29.8	3.1	0.6	8.9	2.0	2.1

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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
White-Tailed Deer												
Adult Female												
31132675209211	0.0	0.0	9.0	1.4	34.9	35.6	9.5	0.3	1.0	4.0	0.1	0.4
31432575209211	0.1	0.0	20.7	2.3	16.0	21.5	24.9	2.7	0.7	5.3	1.0	2.0
31732875208211	0.1	0.0	18.6	1.5	18.0	18.9	25.0	2.1	1.6	5.4	1.1	2.6
32132875209211	0.2	0.0	22.6	2.1	16.2	22.9	18.8	4.3	1.0	6.8	1.6	2.9
33203274209301	0.1	0.0	16.7	1.8	16.4	18.3	30.9	3.9	0.4	7.2	1.6	1.4
33393274309301	0.1	0.0	15.8	1.4	15.8	17.2	34.0	3.4	0.5	7.2	1.7	1.7
34003274309301	0.2	0.0	15.7	1.5	15.3	18.4	31.7	4.1	0.5	7.4	1.7	1.7
91703174308301	0.2	0.1	19.3	2.8	18.3	15.7	26.1	4.1	0.6	7.1	2.2	1.3
99903274309301	0.3	0.2	16.5	2.0	17.3	15.9	30.0	4.8	0.5	6.8	1.7	1.8
Juvenile Male												
90402874105301	0.1	0.0	15.4	1.2	16.8	13.8	34.6	4.5	0.8	6.4	2.5	2.2
Juvenile Female												
41413977206405	0.1	0.0	16.0	1.7	23.8	8.9	28.3	4.9	0.7	7.9	2.1	1.9
41413977206404	0.2	0.2	16.8	2.1	22.0	9.6	27.1	5.4	0.7	7.0	1.8	2.5
41413977206401	0.2	0.1	12.0	1.6	17.0	8.6	30.0	5.9	1.1	11.5	4.1	3.8
41413977206407	0.3	0.1	13.4	1.9	17.8	8.3	28.5	6.2	1.0	11.1	3.6	3.1
41413977206408	0.1	0.2	16.4	1.8	18.1	7.3	27.7	4.9	0.9	11.0	3.5	3.3
41413977206409	0.1	0.1	12.7	1.3	21.7	8.8	28.2	5.5	0.9	11.6	3.3	3.0
41603277205405	0.1	0.0	14.2	2.6	17.1	15.8	25.6	4.1	1.2	10.3	2.9	3.9
41603277205404	0.2	0.1	15.8	2.1	16.6	17.6	26.1	4.2	1.2	8.2	2.4	3.0
41603277205401	0.2	0.1	14.6	2.0	18.0	20.8	22.4	5.0	1.0	8.1	2.4	2.6
41603277205408	0.2	0.1	18.0	3.7	18.0	16.0	24.1	3.3	1.1	9.3	2.0	3.1
41603277205409	0.1	0.0	15.1	3.0	20.1	15.6	24.5	3.2	1.0	9.2	2.5	3.2
91103074205301	0.2	0.0	14.4	1.4	15.8	12.1	34.5	4.5	0.7	8.4	3.8	2.5

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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
White-Tailed Deer												
Immature Male												
31032675104211	0.2	0.0	26.1	2.2	20.4	14.9	25.6	2.1	0.7	4.1	0.8	1.3
32432675104211	0.4	0.1	19.1	2.3	22.6	15.6	22.0	3.3	0.7	6.0	2.1	3.0
41332677104301	0.2	0.0	15.8	3.2	17.0	12.0	23.3	6.5	0.9	6.8	5.7	5.1
Immature Female												
31532675204211	0.1	0.0	19.9	2.2	16.9	16.8	22.5	2.2	1.5	9.0	2.6	4.0
32232575204211	0.6	0.1	19.2	1.6	17.1	16.9	22.5	2.4	0.9	6.9	2.2	3.4
Mule Deer												
Adult Male												
21911676109301	1.5	0.6	13.7	2.2	17.4	17.4	21.4	6.4	0.7	8.2	3.2	2.9
30032675109211	0.1	0.0	15.8	2.6	17.9	20.9	17.8	3.3	1.2	5.2	1.1	1.8
30533275109211	0.1	0.0	17.8	2.7	16.2	25.1	20.6	3.8	1.1	6.4	1.3	1.7
30632975109211	0.1	0.0	21.5	1.7	21.2	15.2	20.5	4.8	0.7	7.7	1.8	2.4
30833275109211	0.1	0.0	18.2	2.1	20.5	32.3	33.5	4.5	1.7	8.4	1.6	2.3
Adult Female												
22332575209301	0.2	0.3	17.3	2.9	17.2	16.4	21.2	6.5	0.7	7.8	3.2	2.8
22818175209301	0.1	0.0	8.0	1.2	18.5	15.9	25.7	5.2	1.2	10.3	4.4	3.6
30333075209211	0.1	0.0	18.9	1.4	17.2	20.4	20.6	4.4	0.6	8.2	3.0	3.3
30433275209211	0.1	0.0	16.3	2.1	21.6	18.6	20.8	2.8	1.5	6.5	2.0	2.9
30333075209301	0.1	0.0	12.7	2.0	15.3	24.5	21.1	5.8	0.6	9.6	3.8	3.2
30433275209301	0.1	0.0	17.0	2.0	21.2	18.2	24.4	3.4	1.1	5.9	1.9	1.8
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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
Mule Deer												
Immature Male												
22033075104301	0.6	0.3	15.8	2.2	19.7	17.8	23.4	5.7	0.6	4.5	3.5	2.5
30132575104211	0.1	0.0	14.4	2.5	16.5	23.6	24.2	5.6	0.6	7.1	2.4	2.6
30232575104211	0.3	0.0	19.1	0.9	21.1	17.0	25.3	5.3	1.0	10.8	2.7	3.0
30732975104211	0.1	0.0	21.9	1.4	19.6	24.0	19.7	3.8	0.5	4.4	1.2	1.3
Immature Female												
30933875204201	0.8	0.0	10.2	2.2	21.6	12.7	30.6	5.0	0.7	6.4	3.2	2.4
Antelope												
Adult Male												
20130075109301	1.6	0.4	16.0	2.1	13.9	14.1	22.9	4.1	1.0	12.3	2.5	2.7
20230075109301	1.5	0.3	17.4	1.0	13.5	9.9	28.8	3.3	0.8	13.2	1.8	1.8
20325376108301	0.2	0.0	15.0	1.2	15.5	18.0	29.2	4.1	0.6	10.4	2.1	2.2
20430076109301	0.5	0.2	16.2	1.2	12.4	11.0	28.3	4.5	1.1	13.2	2.8	2.6
20530076109301	0.5	0.2	17.8	1.5	13.4	11.1	28.4	3.5	1.0	11.1	2.0	1.9
23430076109401	0.2	0.2	15.7	1.9	15.0	15.0	22.1	3.7	0.8	15.0	3.7	3.6
23430076109404	0.1	0.0	11.8	1.4	20.0	21.2	17.9	3.1	0.9	14.2	2.2	3.8
23430076109405	0.2	0.1	13.7	2.1	16.5	16.4	20.0	4.1	0.7	15.0	3.8	4.1
23430076109407	0.1	0.2	16.8	1.9	18.9	15.7	20.1	3.1	0.8	13.8	2.5	2.7
23430076109408	0.2	0.0	14.7	1.0	17.4	16.8	21.8	3.2	0.9	15.3	3.0	2.9
23430076109409	0.1	0.1	13.6	1.7	18.4	16.3	20.6	3.5	0.7	15.5	3.3	4.1
23430076109410	0.1	0.0	14.8	1.6	18.9	15.6	22.5	3.1	0.8	14.1	3.0	2.8
Bighorn Sheep												
Adult Female												
20628076209301	2.4	1.4	13.3	2.7	11.9	19.4	21.4	6.7	0.6	6.7	3.5	2.6

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APPENDIX I (Continued)

Identification No.	14:0	14:1	16:0	16:1	18:0	18:1	18:2	18:3	20:3	20:4	20:5	22:5
Phospholipids												
Bighorn Sheep												
Adult Female												
20728076209301	3.0	1.2	15.3	3.5	12.8	24.1	16.9	5.7	0.4	5.6	3.5	2.3
20827976209301	1.6	0.8	11.9	2.0	11.7	17.7	23.1	8.1	0.6	7.5	4.0	2.8
20928076209301	2.4	1.8	11.7	2.3	10.8	15.4	23.0	8.1	0.4	6.5	3.9	2.7
21027976209206	3.0	1.5	18.8	3.1	19.9	18.8	19.8	3.8	0.2	2.2	0.8	0.5
21126676209301	0.5	0.3	12.9	2.4	11.0	18.1	20.8	6.8	0.5	7.0	3.9	2.6
21226576209301	0.8	0.5	13.4	2.7	10.7	20.6	18.0	7.2	0.4	6.1	3.3	2.3
21627976209206	0.8	0.2	17.4	2.2	14.7	21.2	19.4	5.3	0.6	7.4	3.4	3.0
23507377409401	0.1	0.0	10.5	2.7	9.9	22.1	26.5	7.1	0.5	9.0	4.9	3.6
23507377409407	0.6	0.1	15.4	3.6	13.4	24.8	19.4	4.9	0.4	7.5	3.8	2.2
23507377409408	0.1	0.1	16.0	3.1	14.2	17.7	22.2	4.9	0.6	9.0	4.3	3.6
23507377409409	0.1	0.1	12.9	2.7	14.5	15.2	22.9	5.3	0.6	11.4	5.7	4.7

APPENDIX 2 Classification function coefficients

2A. Species

2B. Age

2C. Sex

Triacylglycerols

	Elk	Moose	W.T.D.	M.D.	Ant.	B.H.S.
14:0	34.63792	29.35069	29.67981	29.61806	31.07779	34.19914
14:1	99.31873	107.86287	107.84169	108.93503	106.29770	101.61609
16:0	44.39664	44.25694	45.12794	45.50832	43.66956	44.09854
16:1	36.58070	32.95987	33.67957	33.66869	33.00230	34.02867
18:0	44.03815	43.17007	43.88339	44.13342	43.08606	43.39310
18:1	42.65495	42.32286	42.86783	43.11978	42.17688	42.83145
18:2	63.65985	63.49937	63.54652	63.34393	62.26460	63.33318
18:3	26.67680	27.21255	29.35127	30.88106	23.79669	29.55186
20:3	248.50768	255.63783	252.45148	252.85017	277.03467	236.19571
20:4	24.46896	20.42867	22.08757	22.71727	21.72537	22.78680
20:5	-90.13454	-83.00632	-75.48154	-80.55736	-63.81458	-71.65125
22:5	303.97461	265.86011	262.53198	277.53149	273.29590	262.85742
Constant	-2119.10938	-2052.03320	-2114.82031	-2141.54492	-2029.46094	-2088.67383

Phospholipids

	Elk	Moose	W.T.D.	M.D.	Ant.	B.H.S.
14:0	17.11780	16.12393	15.15501	15.98120	20.84758	17.72086
14:1	12.33397	11.70250	13.94142	13.22652	7.47933	13.07093
16:0	9.15004	8.92188	9.31479	9.54506	9.47996	9.12676
16:1	15.58265	13.21412	13.33936	13.67497	11.94230	14.10112
18:0	10.85039	10.52252	11.13070	11.57784	10.69971	10.66717
18:1	8.54998	8.65598	8.87624	9.50438	8.86761	9.48683
18:2	7.42535	7.34582	7.31455	7.31771	7.00024	6.90854
18:3	20.01772	19.38319	21.46040	23.45891	19.97850	23.37143
20:3	36.11261	28.16373	32.39009	35.40961	26.45822	29.09180
20:4	10.41331	10.26629	9.45432	9.25812	11.67949	8.96821
20:5	13.28899	12.49439	12.88926	15.37511	12.53170	18.25671
22:5	0.53665	0.43348	2.84567	1.36809	1.15000	-0.49015
Constant	-474.62256	-444.59229	-473.82739	-509.99243	-467.92358	-479.21484

APPENDIX 2B. Classification function coefficients - age.

ELK

		<u>Triacylglycerols</u>	
		<u>7 to 15 months</u>	<u>>15 months</u>
Fatty Acid	14:1	147.14188	149.78397
	16:0	52.77264	54.39355
	16:1	51.84688	54.38278
	18:0	71.44736	73.63948
	18:1	48.14565	49.22487
	18:3	148.85280	156.42743
	20:4	122.81097	129.38503
	Constant	-2428.57617	-2578.16797

		<u>Phospholipids</u>	
		<u>7 to 15 months</u>	<u>>15 months</u>
Fatty Acid	18:2	2.75363	2.18312
	18:3	-1.39212	-0.78524
	20:4	0.98219	1.26429
	Constant	-45.71977	-34.58031

MOOSE

		<u>Triacylglycerols</u>	
		<u>7 to 15 months</u>	<u>>15 months</u>
Fatty Acid	16:0	25.84937	27.00116
	18:0	17.76080	18.92502
	18:1	21.13118	22.81738
	18:2	41.30855	44.84917
	18:3	-30.78444	-38.13940
	20:3	-333.76660	-397.96021
	Constant	-922.15894	-1043.83569

		<u>Phospholipids</u>	
		<u>7 to 15 months</u>	<u>>15 months</u>
Fatty Acid	14:0	34.67241	37.59045
	14:1	229.50050	252.24420
	16:0	16.86626	17.96297
	18:0	17.92151	19.10994
	18:1	17.47882	18.15260
	18:2	15.65748	16.82491
	20:4	40.88324	43.69403
	22:5	-26.01544	-29.08185
	Constant	-789.60620	-892.67676

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APPENDIX 2B (Continued)

WHITE-TAILED DEER

		<u>Triacylglycerols</u>		
		<u>7 to 15 months</u>	<u>>15 months</u>	<u>4 to 6 months</u>
	14:0	136.50444	136.68005	143.71619
	16:0	87.68982	88.65738	88.66054
	16:1	82.89008	82.85028	84.54196
Fatty	18:0	84.20387	84.80719	84.72052
Acid	18:1	85.68364	86.64090	86.70888
	18:2	113.40686	114.30296	111.51477
	20:3	1165.25146	1169.68774	1278.26563
	20:4	47.02745	49.46942	49.88902
	Constant	-4124.34766	-4202.28906	-4215.40625

		<u>Phospholipids</u>		
		<u>7 to 15 months</u>	<u>>15 months</u>	<u>4 to 6 months</u>
	14:1	-73.70049	-85.16336	-93.28415
	16:0	11.90809	11.70725	13.16343
	18:0	11.76436	11.42142	12.71970
	18:2	5.51268	5.24195	5.57785
Fatty	18:3	0.43121	0.41171	-2.19448
Acid	20:3	53.57324	44.97305	42.15619
	20:4	3.56332	3.71801	2.01982
	20:5	13.10693	10.95191	16.46004
	22:5	9.46725	10.57084	16.67088
	Constant	-348.20020	-322.61353	-386.99292

APPENDIX 2C. Classification function coefficients - sex

ELK

		<u>Triacylglycerols</u>	
		Male	Female
Fatty Acid	16:1	2.13555	3.32116
	18:2	2.24591	0.44483
	18:3	8.38470	14.31979
	20:4	5.74578	12.86437
	20:5	-175.79990	-376.05176
	22:5	36.37807	88.40849
	Constant	-19.98718	-36.30647

		<u>Phospholipids</u>	
		Male	Female
Fatty Acid	14:0	-49.04929	-53.82086
	14:1	-31.17284	-26.68752
	16:0	3.72831	3.90305
	16:1	12.15123	14.08965
	18:0	11.78254	12.48804
	18:1	16.63728	17.27710
	20:5	51.64497	54.66731
	Constant	-291.08081	-326.26660

MOOSE

		<u>Triacylglycerols</u>	
		Male	Female
Fatty Acid	14:1	20.25423	17.01192
	16:0	1.21472	1.50563
	18:1	1.54853	1.33232
	20:4	-8.36880	-21.12041
	Constant	-48.55699	-42.87820

		<u>Phospholipids</u>	
		Male	Female
Fatty Acid	14:0	23.02515	18.33408
	16:1	55.42860	48.14568
	18:1	2.97696	3.68427
	18:2	8.36444	7.50023
	18:3	9.23657	3.33602
	20:3	112.16821	76.99800
	20:5	3.94618	10.20313
	22:5	-11.72453	-8.33150
	Constant	-263.28418	-209.98108

APPENDIX 2C (Continued).

WHITE-TAILED DEER

		<u>Triacylglycerols</u>	
		Male	Female
Fatty	14:0	7.55069	4.57911
Acid	18:3	3.18346	2.03727
	Constant	-7.45606	-2.83233

		<u>Phospholipids</u>	
		Male	Female
	18:1	3.90566	4.17286
Fatty	18:2	4.37525	4.91775
Acid	20:3	31.33470	37.17419
	20:4	2.50787	1.83231
	22:5	0.67053	-1.04162
	Constant	-110.90073	-124.78421

APPENDIX 3 Classification probabilities¹

3A. Species

- 1 = Elk
- 2 = Moose
- 3 = White-tailed deer
- 4 = Mule deer
- 5 = Antelope
- 6 = Bighorn sheep

3B. Age

- 2 = 7 to 15 months
- 3 = >15 months
- 4 = fetal

3C. Sex

- 1 = Male
- 2 = Female

¹ The samples are arranged in the same order as in Appendix 1.

APPENDIX 3A. Classification probabilities - Species

Animal Number	Highest Probability			2nd Highest Probability	
	Species	$P(X/G)^1$	$P(G/X)^2$	Species	$P(G/X)$
Triacylglycerols					
Elk (=1)					
400	1	0.323	1.000		
"	1	0.166	0.865	6	0.057
"	1	0.507	1.000		
"	1	0.187	1.000		
"	1	0.547	1.000		
"	1	0.733	1.000		
403	1	0.130	0.973	5	0.025
"	5	0.111	0.354	1	0.275
"	1	0.336	0.946	2	0.026
"	1	0.226	0.983	3	0.013
"	1	0.138	0.510	5	0.210
"	1	0.956	1.000		
770	1	0.329	1.000		
780	1	0.001	1.000		
936	1	0.468	1.000		
401	1	0.963	1.000		
"	1	0.520	1.000		
"	1	0.930	1.000		
"	1	0.739	1.000		
"	1	0.989	1.000		
"	1	0.980	1.000		
402	1	0.971	1.000		
"	1	0.956	1.000		
"	1	0.510	1.000		
"	1	0.772	1.000		
"	1	0.924	1.000		
"	1	0.270	1.000		
404	1	0.315	1.000		
"	1	0.712	1.000		
"	1	0.816	1.000		
"	1	0.606	1.000		
"	1	0.053	1.000		
"	1	0.008	1.000		
521	1	0.812	1.000		
777	1	0.135	1.000		
778	1	0.826	1.000		
782	1	0.231	1.000		
926	1	0.946	1.000		
927	1	0.468	1.000		

. . . Continued

¹ probability of a case in that group being as far from the centroid as the case in question

² probability of group membership on which the classification was based.

APPENDIX 3A (Continued)

Animal Number	Highest Probability			2nd Highest Probability	
	Species	P(X/G)	P(G/X)	Species	P(G/X)
941	1	0.285	1.000		
408	1	0.991	1.000		
"	1	0.992	1.000		
"	1	0.446	1.000		
"	1	0.971	1.000		
757	2	0.629	0.605	5	0.272
407	1	0.493	0.999		
"	1	0.106	0.937	6	0.057
"	1	0.794	0.998	6	0.001
"	1	0.345	0.914	6	0.041
"	1	0.207	0.996	5	0.003
759	3	0.066	0.487	1	0.351
Moose (=2)					
002	6	0.543	0.621	2	0.291
003	2	0.885	0.656	3	0.161
218	2	0.499	0.599	5	0.201
405	5	0.846	0.473	2	0.408
406	2	0.960	0.889	3	0.055
"	2	0.962	0.885	3	0.086
"	2	0.256	0.758	6	0.213
"	2	0.706	0.816	6	0.082
"	2	0.815	0.939	5	0.038
"	2	0.373	0.545	6	0.398
518	2	0.952	0.690	3	0.168
751	2	0.965	0.769	5	0.144
915	3	0.947	0.536	2	0.251
923	2	0.983	0.726	3	0.172
420	5	0.803	0.474	2	0.433
417	2	0.772	0.855	3	0.098
028	3	0.576	0.587	4	0.234
224	3	0.927	0.697	4	0.174
527	3	0.653	0.317	2	0.302
530	2	0.844	0.550	3	0.332
743	2	0.876	0.425	3	0.392
749	3	0.819	0.476	2	0.296
784	2	0.913	0.682	3	0.142
907	3	0.838	0.437	2	0.340
930	6	0.426	0.460	3	0.254
224	1	0.083	0.652	2	0.277
"	2	0.016	0.823	5	0.177
"	2	0.516	0.938	3	0.060
"	2	0.157	0.997	3	0.002
"	2	0.969	0.865	3	0.089
"	2	0.000	0.999	3	0.001
"	2	0.378	0.487	3	0.301

. . . Continued

APPENDIX 3A (Continued)

Animal Number	Highest Probability			2nd Highest Probability	
	Species	P(X/G)	P(G/X)	Species	P(G/X)
418	3	0.977	0.520	2	0.333
773	2	0.943	0.636	3	0.276
932	2	0.853	0.726	5	0.245
409	2	0.250	0.899	3	0.093
"	2	0.477	0.959	3	0.035
"	2	0.589	0.954	3	0.038
"	2	0.764	0.925	3	0.065
"	2	0.390	0.874	3	0.078
411	2	0.995	0.865	5	0.070
"	2	0.939	0.962	3	0.031
"	2	0.908	0.460	3	0.430
"	2	0.841	0.930	5	0.044

White-tailed deer (=3)

026	3	0.880	0.514	4	0.293
996	3	0.742	0.560	4	0.397
998	3	0.542	0.531	5	0.238
312	4	0.977	0.750	3	0.244
313	4	0.865	0.651	3	0.322
316	4	0.989	0.646	3	0.332
318	2	0.543	0.420	3	0.301
319	4	0.980	0.635	3	0.348
320	4	0.919	0.735	3	0.263
323	4	0.936	0.581	3	0.385
412	6	0.145	0.564	3	0.285
"	3	0.000	0.847	6	0.137
"	3	0.021	0.847	4	0.053
"	3	0.608	0.854	4	0.087
"	3	0.527	0.743	2	0.165
"	3	0.798	0.653	4	0.300
415	2	0.890	0.765	3	0.206
"	3	0.993	0.677	4	0.233
"	2	0.903	0.776	3	0.203
"	2	0.751	0.823	6	0.090
229	3	0.947	0.472	4	0.423
332	3	0.897	0.491	2	0.251
333	3	0.974	0.601	4	0.278
340	3	0.975	0.543	4	0.313
917	2	0.798	0.485	3	0.299
999	3	0.926	0.509	4	0.445
311	3	0.580	0.388	4	0.286
314	4	0.957	0.722	3	0.272
317	4	0.811	0.644	3	0.349
321	4	0.689	0.814	3	0.184
904	3	0.987	0.556	4	0.230
418	3	0.701	0.528	4	0.432

. . . Continued

APPENDIX 3A (Continued)

Animal Number	Highest Probability			2nd Highest Probability	
	Species	P(X/G)	P(G/X)	Species	P(G/X)
419	3	0.957	0.564	4	0.211
028	3	0.992	0.532	4	0.382
033	3	0.694	0.472	4	0.442
911	3	0.682	0.546	2	0.177
414	4	0.718	0.745	3	0.252
"	3	0.000	0.714	5	0.280
"	3	0.069	0.567	4	0.421
"	4	0.665	0.594	3	0.373
"	4	0.456	0.629	3	0.351
416	2	0.850	0.811	3	0.167
"	3	0.799	0.578	2	0.312
"	3	0.040	0.810	5	0.094
"	2	0.980	0.648	3	0.308
"	2	0.981	0.872	3	0.106
226	3	0.988	0.557	4	0.377
"	3	0.719	0.595	4	0.381
"	3	0.605	0.597	4	0.363
"	3	0.355	0.535	4	0.433
"	3	0.974	0.558	4	0.274
"	3	0.164	0.386	6	0.329
310	3	0.739	0.409	4	0.364
324	6	0.675	0.891	4	0.061
413	3	0.996	0.679	4	0.217
315	6	0.784	0.886	5	0.088
322	6	0.649	0.643	4	0.186

Mule Deer (=4)

219	4	0.456	0.365	3	0.314
300	4	0.962	0.756	3	0.240
305	4	0.892	0.651	3	0.337
306	4	0.990	0.782	3	0.213
308	4	0.676	0.794	3	0.203
221	3	0.973	0.495	4	0.428
222	4	0.789	0.793	3	0.206
223	2	0.197	0.325	4	0.278
228	4	0.447	0.685	3	0.302
303	4	0.955	0.830	3	0.168
304	4	0.692	0.755	3	0.244
227	4	0.408	0.802	3	0.194
220	6	0.888	0.944	3	0.023
301	4	0.961	0.807	3	0.191
302	4	0.890	0.687	3	0.302
307	4	0.998	0.566	3	0.415
309	4	0.994	0.651	3	0.339

. . . Continued

APPENDIX 3A (Continued)

Animal Number	Highest Probability		2nd Highest Probability		
	Species	P(X/G)	P(G/X)	Species	P(G/X)
Antelope (=5)					
201	5	0.003	1.000		
202	5	0.058	1.000		
203	5	0.963	0.938	2	0.032
204	5	0.873	0.989	2	0.009
205	3	0.543	0.414	4	0.317
234	5	0.790	0.701	2	0.293
"	3	0.703	0.332	2	0.320
"	2	0.798	0.648	5	0.247
"	5	0.957	0.794	2	0.131
"	5	0.670	0.975	2	0.012
"	5	0.592	0.985	2	0.015
"	5	0.565	0.324	3	0.321
Bighorn Sheep (=6)					
206	6	0.991	0.998	3	0.001
207	6	0.895	0.952	3	0.018
208	6	0.955	0.997	5	0.002
209	6	0.773	0.996	5	0.001
210	6	0.848	0.999	2	0.001
211	6	0.980	0.993	3	0.003
212	6	0.961	0.990	3	0.004
216	6	0.791	0.992	4	0.004
235	6	0.438	0.585	2	0.338
"	6	0.676	0.955	2	0.037
"	6	0.768	0.938	2	0.044
"	6	0.000	0.996	5	0.004
Phospholipids					
Elk (=1)					
400	1	0.901	0.986	2	0.012
"	1	0.648	0.430	3	0.347
"	2	0.335	0.604	1	0.306
"	1	0.430	0.862	2	0.052
"	2	0.199	0.551	1	0.405
"	2	0.601	0.739	1	0.148
403	1	0.314	0.977	5	0.022
"	1	0.337	0.966	2	0.029
"	1	0.188	0.577	5	0.414
"	2	0.724	0.551	1	0.259
"	1	0.754	0.987	3	0.007
"	5	0.171	0.689	1	0.301
770	1	0.158	1.000		

. . . Continued

APPENDIX 3A (Continued)

Animal Number	Highest Probability			2nd Highest Probability	
	Species	P(X/G)	P(G/X)	Species	P(G/X)
780	1	0.275	0.996	2	0.004
936	1	0.407	0.789	3	0.133
401	1	0.594	0.972	2	0.028
"	1	0.159	1.000		
"	1	0.471	0.837	5	0.107
"	1	0.063	0.978	5	0.018
"	1	0.251	1.000		
"	1	0.139	0.996	5	0.004
402	1	0.583	0.874	3	0.077
"	1	0.984	0.989	2	0.009
"	1	0.963	0.968	2	0.016
"	1	0.745	0.983	2	0.010
"	1	0.891	0.973	2	0.017
"	1	0.830	0.972	2	0.020
404	1	0.583	1.000		
"	1	0.241	1.000		
"	1	0.524	1.000		
"	1	0.137	1.000		
"	1	0.568	1.000		
"	1	0.669	0.999	2	0.001
408	2	0.443	0.336	5	0.330
"	1	0.969	0.933	2	0.050
"	1	0.562	1.000		
"	1	0.440	0.999	3	0.001
"	3	0.788	0.931	4	0.061
521	1	0.875	0.988	3	0.006
777	1	0.890	0.655	2	0.263
778	1	0.751	0.842	2	0.107
926	1	0.674	0.992	3	0.006
927	1	0.216	0.996	3	0.004
407	1	0.609	0.870	2	0.083
"	1	0.765	0.717	2	0.192
"	1	0.706	0.972	2	0.020
"	1	0.375	0.813	2	0.142
"	1	0.780	0.892	2	0.057
757	1	0.786	0.938	2	0.059
759	1	0.840	0.855	2	0.120
Moose (=2)					
218	5	0.040	0.838	6	0.157
405	2	0.316	0.915	5	0.068
406	2	0.838	0.787	1	0.180
"	2	0.881	0.931	1	0.061
"	2	0.641	0.736	1	0.135
"	1	0.888	0.701	2	0.280
"	2	0.716	0.637	1	0.349

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APPENDIX 3A (Continued)

Animal Number	Highest Probability			2nd Highest Probability	
	Species	P(X/G)	P(G/X)	Species	P(G/X)
406	2	0.772	0.756	1	0.199
518	2	0.219	0.993	1	0.005
915	2	0.297	0.998	3	0.002
923	2	0.524	0.994	1	0.006
417	2	0.159	0.990	1	0.010
028	2	0.340	0.537	3	0.454
038	2	0.773	0.851	3	0.128
224	2	0.415	0.935	5	0.051
"	2	0.738	0.875	3	0.085
"	2	0.440	0.846	3	0.150
"	2	0.921	0.983	5	0.008
"	2	0.921	0.989	3	0.006
"	2	0.879	0.993	5	0.004
"	2	0.846	0.861	5	0.091
"	2	0.832	0.996	5	0.003
527	2	0.904	0.818	3	0.094
530	2	0.849	0.830	3	0.165
784	2	0.818	0.753	3	0.236
418	2	0.606	0.990	5	0.005
773	2	0.856	0.955	3	0.044
409	2	0.527	0.589	3	0.219
"	2	0.658	0.440	3	0.276
"	1	0.735	0.933	2	0.046
"	1	0.140	0.506	2	0.486
"	2	0.833	0.623	5	0.241
932	2	0.945	0.950	3	0.041
411	2	0.676	0.558	3	0.428
"	2	0.832	0.962	3	0.037
"	2	0.909	0.976	1	0.018
"	2	0.562	0.746	3	0.252

White-tailed Deer (=3)

312	4	0.799	0.676	3	0.318
313	3	0.919	0.953	2	0.034
316	3	0.459	0.489	5	0.256
318	4	0.113	0.378	3	0.374
319	3	0.830	0.861	2	0.088
323	3	0.718	0.891	4	0.072
412	3	0.560	0.959	4	0.039
"	4	0.841	0.607	3	0.387
"	3	0.943	0.864	4	0.071
"	3	0.850	0.950	4	0.038
"	3	0.699	0.955	4	0.044
"	3	0.761	0.958	4	0.040
320	3	0.209	0.913	2	0.047
415	2	0.434	0.485	1	0.223

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APPENDIX 3A (Continued)

Animal Number	Highest Probability			2nd Highest Probability	
	Species	P(X/G)	P(G/X)	Species	P(G/X)
415	3	0.796	0.726	2	0.239
"	2	0.660	0.546	1	0.276
"	2	0.581	0.444	5	0.410
996	2	0.814	0.602	3	0.339
998	2	0.828	0.759	3	0.193
311	4	0.001	0.999	3	0.001
314	3	0.875	0.887	2	0.093
317	3	0.256	0.984	4	0.012
321	4	0.557	0.827	3	0.172
332	2	0.545	0.819	3	0.173
333	2	0.619	0.924	3	0.075
340	2	0.561	0.781	3	0.210
917	3	0.517	0.537	4	0.360
999	3	0.798	0.843	2	0.083
904	3	0.499	0.891	4	0.058
414	3	0.912	0.931	4	0.060
"	3	0.443	0.964	4	0.035
"	3	0.823	0.880	4	0.094
"	3	0.785	0.771	4	0.190
"	3	0.799	0.940	1	0.033
"	3	0.946	0.890	4	0.092
416	3	0.837	0.927	4	0.044
"	3	0.925	0.769	4	0.226
"	4	1.000	0.936	3	0.063
"	3	0.544	0.701	1	0.277
"	3	0.896	0.905	1	0.054
911	3	0.454	0.601	2	0.325
310	3	0.739	0.962	2	0.031
324	3	0.661	0.920	4	0.080
413	4	0.000	0.648	6	0.347
315	3	0.314	0.969	1	0.021
322	3	0.532	0.970	2	0.021
Mule Deer (=4)					
219	6	0.357	0.513	4	0.480
300	3	0.796	0.745	4	0.246
305	4	0.905	0.914	3	0.085
306	3	0.363	0.585	4	0.414
308	4	0.000	1.000		
223	4	0.635	0.929	6	0.057
228	4	0.937	0.884	3	0.106
303	4	0.282	0.703	3	0.289
304	3	0.273	0.663	4	0.336
303	6	0.339	0.805	4	0.195
304	4	0.611	0.625	3	0.373
220	4	0.266	0.865	6	0.132

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APPENDIX 3A (Continued)

Animal Number	Highest Probability			2nd Highest Probability	
	Species	P(X/G)	P(G/X)	Species	P(G/X)
301	4	0.843	0.958	6	0.026
302	4	0.066	0.938	3	0.060
307	4	0.457	0.933	3	0.067
309	4	0.451	0.635	3	0.357
Antelope (=5)					
201	5	0.811	0.997	2	0.002
202	5	0.233	0.998	2	0.002
203	2	0.628	0.720	5	0.195
204	5	0.915	0.840	2	0.130
205	2	0.811	0.753	5	0.190
234	5	0.978	0.997	2	0.002
"	5	0.630	0.997	3	0.002
"	5	0.820	0.998	3	0.001
"	5	0.995	0.991	2	0.007
"	5	0.798	1.000		
"	5	0.870	0.999	2	0.001
"	5	0.990	0.991	2	0.008
Bighorn Sheep (=6)					
206	6	0.975	1.000		
207	6	0.232	1.000		
208	6	0.949	1.000		
209	6	0.439	1.000		
210	6	0.495	0.782	4	0.128
211	6	0.915	1.000		
212	6	0.859	1.000		
216	6	0.568	0.871	4	0.127
235	6	0.823	1.000		
"	6	0.434	1.000		
"	6	0.653	0.867	4	0.105
"	6	0.629	0.976	4	0.018

APPENDIX 3B. Classification probabilities - Age

Animal Number	Actual Age	Highest Probability		2nd Highest Probability		
		Age	P(X/G)	P(G/X)	Age	P(G/X)
Triacylglycerols						
Elk						
400	3	3	0.997	1.000		
"		3	1.000	0.975	2	0.025
"		2	0.986	0.775	3	0.225
"		3	0.997	1.000		
"		3	0.961	0.626	2	0.374
"		2	0.987	0.785	3	0.215
403	3	3	0.988	0.789	2	0.211
"		3	0.994	0.853	2	0.147
"		2	0.949	0.574	3	0.426
"		3	1.000	0.993	2	0.007
"		2	0.934	0.517	3	0.483
"		3	0.995	0.866	2	0.134
770	3	3	0.999	0.933	2	0.067
780	3	2	0.944	0.552	3	0.448
936	3	3	1.000	0.998	2	0.002
401	3	3	1.000	0.991	2	0.009
"		3	1.000	0.988	2	0.012
"		3	1.000	0.987	2	0.013
"		3	1.000	0.982	2	0.018
"		3	0.999	0.999	2	0.001
"		3	0.999	0.999	2	0.001
402	3	3	1.000	0.997	2	0.003
"		3	1.000	0.997	2	0.003
"		3	0.999	0.999	2	0.001
"		3	0.989	1.000		
"		3	1.000	0.999	2	0.001
"		3	0.999	0.999	2	0.001
404	3	3	0.999	0.918	2	0.082
"		2	0.988	0.793	3	0.207
"		3	0.999	0.999	2	0.001
"		3	0.967	1.000		
"		3	0.993	1.000		
"		3	1.000	0.991	2	0.009
521	3	3	1.000	0.997	2	0.003
777	3	3	0.683	1.000		
778	3	3	0.998	0.999	2	0.001
782	3	3	0.999	0.999	2	0.001
926	3	3	1.000	0.998	2	0.002
927	3	3	1.000	0.996	2	0.004
941	3	3	1.000	0.996	2	0.004
408	3	3	1.000	0.994	2	0.006

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APPENDIX 3B (Continued)

Animal Number	Actual Age	Highest Probability		2nd Highest Probability	
		Age	P(X/G)	P(G/X)	Age
408	3	3	1.000	0.990	2
"		3	0.998	1.000	
"		3	1.000	0.997	2
757	2	2	1.000	0.999	3
407	2	2	0.997	0.891	3
"		2	0.918	1.000	
"		2	0.999	0.921	3
"		2	1.000	0.986	3
"		2	1.000	0.984	3
759	2	2	1.000	0.992	3
Moose					
002	3	3	0.952	1.000	
003	3	3	0.480	1.000	
218	3	3	0.271	0.973	2
405	3	3	0.626	0.998	2
406	3	3	0.547	1.000	
"		2	0.064	0.645	3
"		3	0.245	1.000	
"		3	0.239	1.000	
"		3	0.815	1.000	
"		3	0.168	1.000	
518	3	3	0.545	1.000	
751	3	3	0.951	1.000	
915	3	3	0.064	0.642	2
923	3	3	0.943	1.000	
420	3	3	0.096	1.000	
417	3	3	0.103	0.814	2
028	3	3	0.749	1.000	
224	3	3	0.635	1.000	
527	3	3	0.383	1.000	
530	3	3	0.472	1.000	
743	3	3	0.996	1.000	
749	3	3	0.735	0.999	2
784	3	3	0.632	1.000	
907	3	3	0.744	0.999	2
930	3	3	0.906	0.999	2
224	3	2	0.096	0.792	3
"		3	0.059	0.611	2
"		3	0.408	1.000	
"		3	0.908	0.999	2
"		3	0.510	1.000	
"		3	0.793	1.000	
"		3	0.519	0.996	2
418	3	3	0.443	1.000	
773	2	2	0.133	0.879	3

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APPENDIX 3B (Continued)

Animal Number	Actual Age	Highest Probability			2nd Highest Probability	
		Age	P(X/G)	P(G/X)	Age	P(G/X)
932	2	2	0.702	0.998	3	0.002
409	2	2	0.188	1.000		
"		2	0.566	1.000		
"		2	0.937	1.000		
"		2	0.486	0.995	3	0.005
"		2	0.173	1.000		
411	2	2	0.412	0.991	3	0.009
"		2	0.719	0.999	3	0.001
"		2	0.909	1.000		
"		2	0.637	1.000		

White-Tailed Deer

026	3	3	0.585	0.679	2	0.321
996	3	2	0.855	0.643	3	0.357
998	3	3	0.663	0.505	2	0.495
312	3	3	0.348	0.945	2	0.051
313	3	3	0.967	0.778	2	0.222
316	3	3	0.974	0.741	2	0.259
318	3	3	0.289	0.920	2	0.068
319	3	3	0.359	0.976	2	0.023
320	3	3	0.961	0.844	2	0.156
323	3	3	0.545	0.783	2	0.209
412	3	3	0.720	0.582	2	0.418
"		3	0.708	0.793	2	0.207
"		3	0.208	0.988	2	0.012
"		3	0.786	0.736	2	0.264
"		3	0.746	0.579	2	0.421
"		2	0.934	0.698	3	0.302
415	3	2	0.950	0.748	3	0.252
"		4	0.445	0.995	2	0.005
"		2	0.289	0.536	3	0.464
"		3	0.823	0.636	2	0.363
229	3	3	0.596	0.959	2	0.041
332	3	3	0.448	0.560	2	0.440
333	3	3	0.614	0.907	2	0.093
340	3	3	0.871	0.907	2	0.093
917	3	3	0.700	0.943	2	0.057
999	3	3	0.703	0.600	2	0.400
311	3	3	0.394	0.956	2	0.044
314	3	3	0.360	0.978	2	0.022
317	3	3	0.497	0.966	2	0.034
321	3	3	0.359	0.979	2	0.021
904	2	3	0.819	0.684	2	0.315
418	2	2	0.873	0.873	3	0.127
419	2	2	0.313	0.646	3	0.354
028	2	2	0.619	0.782	3	0.207

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APPENDIX 3B (Continued)

Animal Number	Actual	Highest Probability		2nd Highest Probability		
	Age	Age	P(X/G)	P(G/X)	Age	P(G/X)
033	2	2	0.320	0.860	3	0.085
911	2	2	0.107	0.807	4	0.166
414	2	2	0.507	0.757	3	0.222
"		2	0.765	0.702	3	0.298
"		2	0.281	0.971	3	0.029
"		2	0.440	0.952	3	0.048
"		2	0.042	0.993	3	0.007
416	2	3	0.561	0.503	2	0.495
"		2	0.862	0.895	3	0.104
"		2	0.898	0.660	3	0.340
"		3	0.879	0.811	2	0.189
"		3	0.834	0.667	2	0.333
226	4	4	0.803	0.996	2	0.002
"		4	0.752	1.000		
"		4	0.115	1.000		
"		2	0.164	0.629	4	0.280
"		4	0.642	0.991	2	0.008
"		4	0.314	1.000		
310	4	4	0.789	0.994	2	0.004
324	4	4	0.750	1.000		
413	4	4	0.589	0.985	2	0.012
315	4	4	0.956	1.000		
322	4	4	0.047	1.000		

Phospholipids

Elk

400	3	2	0.539	0.637	3	0.363
"		3	0.870	0.801	2	0.199
"		3	0.934	0.864	2	0.136
"		3	0.582	0.664	2	0.336
"		3	0.823	0.783	2	0.217
"		3	0.903	0.872	2	0.128
403	3	3	0.587	0.937	2	0.063
"		3	0.654	0.704	2	0.296
"		3	0.081	0.993	2	0.007
"		3	0.818	0.781	2	0.219
"		3	0.288	0.975	2	0.025
"		3	0.259	0.978	2	0.022
770	3	2	0.619	0.685	3	0.315
780	3	3	0.381	0.521	2	0.479
936	3	2	0.760	0.905	3	0.095
401	3	3	0.592	0.670	2	0.330
"		3	0.361	0.503	2	0.497
"		3	0.449	0.956	2	0.044

. . . Continued

APPENDIX 3B (Continued)

Animal Number	Actual	Highest Probability		2nd Highest Probability		
	Age	Age	P(X/G)	P(G/X)	Age	P(G/X)
401	3	3	0.809	0.894	2	0.106
"		3	0.800	0.774	2	0.226
"		3	0.338	0.969	2	0.031
402	3	3	0.593	0.670	2	0.330
"		3	0.711	0.915	2	0.085
"		3	0.698	0.917	2	0.083
"		3	0.457	0.955	2	0.045
"		3	0.572	0.939	2	0.061
"		3	0.708	0.915	2	0.085
404	3	3	0.907	0.871	2	0.129
"		3	0.734	0.911	2	0.089
"		3	0.501	0.949	2	0.051
"		3	0.224	0.981	2	0.019
"		3	0.868	0.881	2	0.119
"		3	0.304	0.973	2	0.027
408	3	3	0.052	0.995	2	0.005
"		3	0.485	0.952	2	0.048
"		3	0.256	0.978	2	0.022
"		3	0.776	0.763	2	0.237
"		3	0.967	0.855	2	0.145
521	3	2	0.803	0.775	3	0.225
777	3	2	0.782	0.901	3	0.099
778	3	3	0.429	0.559	2	0.441
926	3	2	0.851	0.885	3	0.115
927	3	3	0.654	0.925	2	0.075
407	2	2	0.881	0.805	3	0.195
"		3	0.605	0.677	2	0.323
"		2	0.489	0.604	3	0.396
"		2	0.472	0.953	3	0.047
"		3	0.616	0.683	2	0.317
757	2	2	0.181	0.985	3	0.015
759	2	2	0.148	0.987	3	0.013
Moose						
218	3	3	0.930	0.989	2	0.011
405	3	3	0.803	0.993	2	0.007
406	3	3	0.856	0.977	2	0.023
"		3	0.544	0.924	2	0.076
"		3	0.857	0.992	2	0.008
"		3	0.610	0.997	2	0.003
"		3	0.688	0.996	2	0.004
"		3	0.950	0.988	2	0.012
518	3	3	0.359	0.999	2	0.001
915	3	3	0.664	0.953	2	0.047
923	3	3	0.041	1.000		

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APPENDIX 3B (Continued)

Animal Number	Actual Age	Highest Probability		2nd Highest Probability		
		Age	P(X/G)	P(G/X)	Age	P(G/X)
417	3	3	0.305	0.781	2	0.219
028	3	3	0.202	1.000		
038	3	3	0.600	0.939	2	0.061
224	3	3	0.826	0.993	2	0.007
"		3	0.180	1.000		
"		3	0.470	0.896	2	0.104
"		3	0.752	0.994	2	0.006
"		3	0.271	0.742	2	0.258
"		3	0.359	0.999	2	0.001
"		3	0.493	0.906	2	0.094
"		3	0.914	0.990	2	0.010
527	3	3	0.401	0.860	2	0.140
530	3	3	0.287	0.761	2	0.239
784	3	2	0.263	0.730	3	0.270
418	3	3	0.719	0.995	2	0.005
773	2	2	0.464	0.894	3	0.106
409	2	2	0.543	0.998	3	0.002
"		2	0.593	0.997	3	0.003
"		2	0.797	0.993	3	0.007
"		2	0.002	1.000		
"		2	0.587	0.936	3	0.064
932	2	2	0.180	0.586	3	0.414
411	2	2	0.545	0.998	3	0.002
"		2	0.257	0.722	3	0.278
"		2	0.520	0.916	3	0.084
"		2	0.462	0.893	3	0.107

White-Tailed Deer

312	3	3	0.673	0.974	2	0.026
313	3	3	0.423	0.932	2	0.061
316	3	3	0.254	0.992	2	0.008
318	3	3	0.057	0.997	2	0.003
319	3	3	0.667	0.951	2	0.049
323	3	3	0.713	0.881	2	0.117
412	3	3	0.838	0.886	2	0.114
"		2	0.876	0.928	3	0.071
"		3	0.992	0.889	2	0.111
"		2	0.484	0.590	3	0.410
"		2	0.602	0.578	3	0.482
"		3	0.582	0.772	2	0.228
320	3	3	0.038	0.970	4	0.024
415	3	3	0.778	0.899	2	0.100
"		3	0.200	0.727	2	0.159
"		3	0.863	0.757	2	0.243
"		3	0.762	0.948	2	0.052

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APPENDIX 3B (Continued)

Animal Number	Actual	Highest Probability			2nd Highest Probability	
	Age	Age	P(X/G)	P(G/X)	Age	P(G/X)
996	3	3	0.483	0.843	2	0.157
998	3	2	0.670	0.616	3	0.383
311	3	3	0.947	0.814	2	0.186
314	3	3	0.229	0.978	2	0.018
317	3	2	0.558	0.971	3	0.028
321	3	3	0.252	0.892	2	0.072
332	3	3	0.403	0.981	2	0.019
333	3	3	0.804	0.940	2	0.060
340	3	3	0.313	0.973	2	0.027
917	3	2	0.669	0.556	3	0.444
999	3	3	0.329	0.712	2	0.288
904	2	2	0.964	0.828	3	0.172
414	2	2	0.756	0.683	3	0.316
"		2	0.907	0.754	3	0.245
"		2	0.369	0.987	3	0.013
"		2	0.228	0.968	3	0.032
"		2	0.605	0.976	3	0.023
"		2	0.634	0.954	3	0.046
416	2	2	0.669	0.691	3	0.306
"		2	0.830	0.923	3	0.077
"		2	0.288	0.617	3	0.383
"		2	0.721	0.699	3	0.300
"		2	0.183	0.469	3	0.438
911	2	2	0.698	0.905	3	0.091
310	4	4	0.675	0.998	3	0.001
324	4	4	0.791	1.000		
413	4	4	0.050	1.000		
315	4	4	0.367	0.989	2	0.011
322	4	4	0.482	0.989	3	0.008

APPENDIX 3C. Classification probabilities - Sex

Animal Number	Actual Sex	Highest Probability		2nd Highest Probability		
		Sex	P(X/G)	P(G/X)	Sex	P(G/X)
Triacylglycerols						
Elk						
400	1	1	0.752	0.997	2	0.003
"		1	0.478	0.939	2	0.061
"		1	0.645	0.971	2	0.029
"		1	0.144	1.000		
"		1	0.519	0.949	2	0.051
"		1	0.885	0.989	2	0.011
403	1	1	0.129	1.000		
"		1	0.486	0.999	2	0.001
"		1	0.485	0.999	2	0.001
"		1	0.615	0.999	2	0.001
"		1	0.208	1.000		
"		1	0.621	0.968	2	0.032
770	1	1	0.479	0.939	2	0.061
780	1	1	0.511	0.948	2	0.052
936	1	2	0.601	0.965	1	0.035
401	2	1	0.157	0.624	2	0.376
"		1	0.231	0.767	2	0.233
"		2	0.520	0.950	1	0.050
"		2	0.646	0.998	1	0.002
"		2	0.868	0.996	1	0.004
"		2	0.600	0.999	1	0.001
402	2	2	0.711	0.978	1	0.022
"		2	0.733	0.980	1	0.020
"		2	0.122	1.000		
"		2	0.375	1.000		
"		2	0.474	0.999	1	0.001
"		2	0.174	1.000		
404	2	2	0.743	0.981	1	0.019
"		1	0.350	0.883	2	0.117
"		2	0.381	1.000		
"		2	0.570	0.999	1	0.001
"		2	0.379	1.000		
"		2	0.197	1.000		
521	2	2	0.808	0.985	1	0.015
777	2	2	0.447	0.999	1	0.001
778	2	2	0.668	0.974	1	0.026
782	2	2	0.551	0.956	1	0.044
926	2	2	0.808	0.985	1	0.015
927	2	2	0.525	0.951	1	0.049
941	2	2	0.297	0.842	1	0.158
408	2	2	0.707	0.998	1	0.002

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APPENDIX 3C (Continued)

Animal Number	Actual Sex	Highest Probability		2nd Highest Probability		
		Sex	P(X/G)	P(G/X)	Sex	P(G/X)
408	2	2	0.793	0.997	1	0.003
"		2	0.706	0.978	1	0.022
"		2	0.627	0.998	1	0.002
Moose						
002	1	1	0.766	0.922	2	0.078
003	1	2	0.384	0.550	1	0.450
218	1	1	0.481	0.963	2	0.037
405	1	1	0.594	0.949	2	0.051
406	1	1	0.460	0.612	2	0.388
"		1	0.639	0.943	2	0.057
"		1	0.196	0.988	2	0.012
"		1	0.206	0.987	2	0.013
"		1	0.085	0.995	2	0.005
"		1	0.137	0.992	2	0.008
518	1	2	0.513	0.651	1	0.349
751	1	2	0.581	0.695	1	0.305
915	1	2	0.530	0.958	1	0.042
923	1	2	0.372	0.539	1	0.461
420	1	1	0.879	0.900	2	0.100
417	1	1	0.833	0.909	2	0.091
028	2	2	0.966	0.860	1	0.140
224	2	2	0.181	0.989	1	0.011
527	2	1	0.358	0.527	2	0.473
530	2	2	0.605	0.708	1	0.292
743	2	2	0.641	0.729	1	0.271
749	2	2	0.689	0.936	1	0.064
784	2	2	0.369	0.536	1	0.464
907	2	2	0.799	0.916	1	0.084
930	2	2	0.345	0.514	1	0.486
224	2	2	0.257	0.984	1	0.016
"		2	0.427	0.586	1	0.414
"		2	0.470	0.965	1	0.035
"		2	0.645	0.942	1	0.058
"		2	0.904	0.840	1	0.160
"		2	0.589	0.950	1	0.050
"		2	0.804	0.804	1	0.196
418	2	2	0.834	0.909	1	0.091
White-Tailed Deer						
026	1	2	0.248	0.943	1	0.057
996	1	1	0.608	0.584	2	0.416
998	1	2	0.854	0.696	1	0.304
312	1	1	0.935	0.727	2	0.273
313	1	1	0.892	0.786	2	0.214

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APPENDIX 3C (Continued)

Animal Number	Actual Sex	Highest Probability		2nd Highest Probability	
		Sex	P(X/G)	P(G/X)	Sex
316	1	1	0.515	0.534	2
318	1	1	0.475	0.510	2
319	1	1	0.576	0.567	2
320	1	2	0.666	0.613	1
323	1	1	0.881	0.790	2
412	1	1	0.275	0.938	2
"		1	0.056	0.981	2
"		1	0.820	0.808	2
"		2	0.676	0.618	1
"		1	0.780	0.665	2
"		1	0.358	0.922	2
415	1	1	0.399	0.913	2
"		1	0.010	0.993	2
"		1	0.874	0.704	2
"		1	0.572	0.874	2
229	2	2	0.689	0.845	1
332	2	2	0.877	0.791	1
333	2	2	0.611	0.865	1
340	2	2	0.975	0.759	1
917	2	2	0.805	0.676	1
999	2	2	0.548	0.552	1
311	2	2	0.281	0.937	1
314	2	2	0.600	0.580	1
317	2	2	0.622	0.591	1
321	2	2	0.757	0.655	1
418	1	1	0.560	0.877	2

Phospholipids

Elk

400	1	1	0.647	0.959	2
"		1	0.291	0.988	2
"		1	0.727	0.812	2
"		1	0.415	0.980	2
"		1	0.715	0.807	2
"		1	0.854	0.859	2
403	1	1	0.480	0.671	2
"		1	0.275	0.989	2
"		1	0.326	0.534	2
"		1	0.596	0.965	2
"		1	0.861	0.862	2
"		1	0.799	0.840	2
770	1	1	0.884	0.924	2
780	1	1	0.584	0.740	2

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APPENDIX 3C (Continued)

Animal Number	Actual Sex	Highest Probability		2nd Highest Probability	
		Sex	P(X/G)	P(G/X)	Sex
936	1	1	0.594	0.746	2
401	2	1	0.514	0.696	2
"		1	0.443	0.643	2
"		2	0.458	0.654	1
"		1	0.713	0.806	2
"		2	0.083	0.997	1
"		2	0.577	0.736	1
402	2	2	0.132	0.995	1
"		2	0.611	0.963	1
"		2	0.937	0.884	1
"		2	0.582	0.966	1
"		2	0.485	0.975	1
"		2	0.782	0.941	1
404	2	2	0.469	0.975	1
"		2	0.134	0.995	1
"		2	0.076	0.997	1
"		2	0.222	0.991	1
"		2	0.360	0.984	1
"		2	0.122	0.996	1
408	2	2	0.312	0.519	1
"		1	0.357	0.566	2
"		2	0.739	0.817	1
"		2	0.399	0.605	1
"		1	0.862	0.862	2
521	2	2	0.836	0.853	1
777	2	1	0.541	0.714	2
778	2	2	0.748	0.821	1
926	2	2	0.535	0.710	1
927	2	2	0.402	0.981	1
Moose					
218	1	1	0.929	1.000	
405	1	1	0.674	1.000	
406	1	1	0.965	1.000	
"		1	0.141	1.000	
"		1	0.676	0.999	2
"		1	0.302	1.000	
"		1	0.969	1.000	
"		1	0.185	0.973	2
518	1	1	0.782	1.000	
915	1	1	0.142	0.951	2
923	1	1	0.601	0.999	2
417	1	1	0.360	1.000	
028	2	2	0.340	0.994	1
038	2	2	0.455	1.000	

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APPENDIX 3C (Continued)

Animal Number	Actual Sex	Highest Probability		2nd Highest Probability	
		Sex	P(X/G)	P(G/X)	Sex
224	2	2	0.164	1.000	
"	2	2	0.765	1.000	
"	2	2	0.112	1.000	
"	2	2	0.599	0.999	1
"	2	2	0.460	0.998	1
"	2	2	0.134	0.945	1
"	2	2	0.779	1.000	
"		2	0.853	1.000	
527	2	2	0.211	1.000	
530	2	2	0.518	1.000	
784	2	2	0.063	0.786	1
418	2	2	0.414	0.997	1

White-Tailed Deer

312	1	1	0.310	0.574	2
313	1	2	0.295	0.556	1
316	1	1	0.996	0.932	2
318	1	2	0.724	0.860	1
319	1	1	0.465	0.721	2
323	1	1	0.577	0.980	2
412	1	1	0.158	0.997	2
"		1	0.568	0.788	2
"		1	0.555	0.982	2
"		1	0.850	0.955	2
"		1	0.611	0.978	2
"		1	0.230	0.995	2
320	1	1	0.574	0.980	2
415	1	1	0.200	0.996	2
"		2	0.357	0.626	1
"		1	0.560	0.981	2
"		1	0.109	0.998	2
996	1	1	0.420	0.684	2
998	1	1	0.420	0.685	2
311	2	2	0.789	0.962	1
314	2	2	0.941	0.921	1
317	2	2	0.143	0.997	1
321	2	1	0.559	0.783	2
332	2	2	0.923	0.945	1
333	2	2	0.463	0.987	1
340	2	2	0.787	0.962	1
917	2	2	0.466	0.722	1
999	2	2	0.747	0.868	1

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